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(54) Title: SELECTING ANIMALS FOR PARENTALLY IMPRINTED TRAITS

#### (57) Abstract

The invention relates to methods to select breeding animals or animals destined for slaughter for having desired genotypic or potential phenotypic properties, in particular related to muscle mass and/or fat deposition. The invention provides a method for selecting a pig for having desired genotypic or potential phenotypic properties comprising testing a sample from said pig for the presence of a quantitative trait locus (QTL) located at a Sus scrofa chromosome 2 mapping at position 2p1.7.

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Title: Selecting animals for parentally imprinted traits.

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The invention relates to methods to select breeding animals or animals destined for slaughter for having desired genotypic or potential phenotypic properties, in particular related to muscle mass and/or fat deposition. Breeding schemes for domestic animals have so far focused on farm performance traits and carcass quality. This has resulted in substantial improvements in traits like reproductive success, milk production, lean/fat ratio, prolificacy, growth rate and feed efficiency. Relatively simple performance test data have been the basis for these improvements, and selected traits were assumed to be influenced by a large number of genes, each of small effect (the infinitesimal gene model). There are now some important changes occurring in this area. First, the breeding goal of some breeding organisations has begun to include meat quality attributes in addition to the "traditional" production traits. Secondly, evidence is accumulating that current and new breeding goal traits may involve relatively large effects (known as major genes), as opposed to the infinitesimal model that has been relied on so far.

Modern DNA-technologies provide the opportunity to exploit these major genes, and this approach is a very promising route for the improvement of meat quality, especially since direct meat quality assessment is not viable for potential breeding animals. Also for other traits such as lean/fat ratio, growth rate and feed efficiency, modern DNA technology can be very effective. Also these traits are not always easy to measure in the living animal.

The evidence for several of the major genes originally obtained using segregation analysis, i.e. without any DNA marker information. Afterwards molecular studies were performed to detect the location of these

genes on the genetic map. In practice, and except for alleles of very large effect, DNA studies are required to dissect the genetic nature of most traits of economic importance. DNA markers can be used to localise genes or alleles responsible for qualitative traits like coat colour, and they can also be used to detect genes or alleles with substantial effects on quantitative traits like growth rate, IMF etc. In this case the approach is referred to as QTL (quantitative trait locus) mapping, wherein a QTL comprises at least a part of the nucleic acid genome of an animal where genetic information capable of influencing said quantitative trait (in said animal or in its offspring) is located. Information at DNA level can not only help to fix a specific major gene in a population, but also assist in the selection of a quantitative trait which is already selected for. Molecular information in addition to phenotypic data can increase the accuracy of selection and therefore the selection response.

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Improving meat quality or carcass quality is not just about changing levels of traits like tenderness or marbling, but it is also about increasing uniformity. The existence of major genes provides excellent opportunities for improving meat quality because it allows large steps to be made in the desired direction. Secondly, it will help to reduce variation, since we can fix relevant genes in our products. Another aspect is that selecting for major genes allows differentiation for specific markets. Studies are underway in several species, particularly, pigs, sheep, deer and beef cattle.

In particular, intense selection for meat production has resulted in animals with extreme muscularity and leanness in several livestock species. In recent years it has become feasible to map and clone several of the genes causing these phenotypes, paving the way towards more efficient marker assisted selection, targeted drug development (performance enhancing products) and transgenesis. Mutations in the ryanodine receptor (Fuji

et al, 1991; MacLennan and Phillips, 1993) and myostatin (Grobet et al, 1997; Kambadur et al, 1997; McPherron and Lee, 1997) have been shown to cause muscular hypertrophies in pigs and cattle respectively, while genes with major effects on muscularity and/or fat deposition have for instance been mapped to pig chromosome 4 (Andersson et al, 1994) and sheep chromosome 18 (Cocket et al, 1996).

However, although there have been successes in identifying QTLs, the information is currently of limited use within commercial breeding programmes. Many workers in this field conclude that it is necessary to identify the particular genes underlying the QTL. This is a substantial task, as the QTL region is usually relatively large and may contain many genes. Identification of the relevant genes from the many that may be involved thus remains a significant hurdle in farm animals.

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The invention provides a method for selecting a domestic animal for having desired genotypic or potential phenotypic properties comprising testing said animal for the presence of a parentally imprinted qualitative or quantitative trait locus (QTL). Herein, a domestic animal is defined as an animal being selected or having been derived from an animal having been selected for having desired genotypic or potential phenotypic properties.

Domestic animals provide a rich resource of genetic and phenotypic variation, traditionally domestication involves selecting an animal or its offspring for having desired genotypic or potential phenotypic properties. This selection process has in the past century been facilitated by growing understanding and utilisation of the laws of Mendelian inheritance. One of the major problems in breeding programs of domestic animals is the negative genetic correlation between reproductive capacity and production traits. This is for example the case in cattle (a high milk production generally results

in slim cows and bulls) poultry, broiler lines have a low level of egg production and layers have generally very low muscle growth), pigs (very prolific sows are in general fat and have comparatively less meat) or sheep (high prolific breeds have low carcass quality and vice 5 versa). The invention now provides that knowledge of the parental imprinting character of various traits allows to select for example sire lines homozygous for a paternally imprinted QTL for example linked with muscle production or growth; the selection for such traits can thus be less 10 stringent in dam lines in favour of the reproductive quality. The phenomenon of genetic or parental imprinting has never been utilised in selecting domestic animals, it was never considered feasible to employ this elusive genetic characteristic in practical breeding programmes. 15 The invention provides a breeding programme, wherein knowledge of the parental imprinting character of a desired trait, as demonstrated herein, results in a breeding programme, for example in a BLUP programme, with a modified animal model. This increases the accuracy of 20 the breeding value estimation and speeds up selection compared to conventional breeding programmes. Until now, the effect of a parentally imprinted trait in the estimation of a conventional BLUP programme was neglected; using and understanding the parental character 25 of the desired trait, as provided by the invention, allows selecting on parental imprinting, even without DNA testing. For example, selecting genes characterised by paternal imprinting is provided to help increase uniformity; a (terminal) parent homozygous for the "good 30 or wanted" alleles will pass them to all offspring, regardless of the other parent's alleles, and the offspring will all express the desired parent's alleles. This results in more uniform offspring. Alleles that are interesting or favourable from the maternal side or often 35 the ones that have opposite effects to alleles from the paternal side. For example, in meat animals such as pigs

alleles linked with meat quality traits such as inta-

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muscular fat or muscle mass could be fixed in the dam lines while alleles linked with reduced back fat could be fixed in the sire lines. Other desirable combinations are for example fertility and/or milk yield in the female line with growth rates and/or muscle mass in the male lines.

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In a preferred embodiment, the invention provides a method for selecting a domestic animal for having desired genotypic or potential phenotypic properties comprising 10 testing a nucleic acid sample from said animal for the presence of a parentally imprinted quantitative trait locus (QTL). A nucleic acid sample can in general be obtained from various parts of the animal's body by methods known in the art. Traditional samples for the purpose of nucleic acid testing are blood samples or skin 15 or mucosal surface samples, but samples from other tissues can be used as well, in particular sperm samples, oocyte or embryo samples can be used. In such a sample, the presence and/or sequence of a specific nucleic acid, 20 be it DNA or RNA, can be determined with methods known in the art, such as hybridisation or nucleic acid amplification or sequencing techniques known in the art. The invention provides testing such a sample for the presence of nucleic acid wherein a QTL or allele 25 associated therewith is associated with the phenomenon of parental imprinting, for example where it is determined whether a paternal or maternal allele of said QTL is capable of being predominantly expressed in said animal.

The purpose of breeding programs in livestock is to enhance the performances of animals by improving their genetic composition. In essence this improvement accrues by increasing the frequency of the most favourable alleles for the genes influencing the performance characteristics of interest. These genes are referred to as QTL. Until the beginning of the nineties, genetic improvement was achieved via the use of biometrical methods, but without molecular knowledge of the underlying QTL.

Since the beginning of the nineties and due to recent developments in genomics, it is conceivable to identify the QTL underlying a trait of interest. The invention now provides identifying and using parentally imprinted QTLs which are useful for selecting animals by mapping quantitative trait loci. Again, the phenomenon of genetic or paternal imprinting has never been utilised in selecting domestic animals, it was never considered feasible to employ this elusive genetic characteristic in practical breeding programmes. For example Kovacs and Kloting (Biochem. Mol. Biol. Int. 44:399-405, 1998), where parental imprinting is not mentioned, and not suggested, found linkage of a trait in female rats, but not in males, suggesting a possible sex specificity associated with a chromosomal region, which of course excludes parental imprinting, a phenomenon wherein the imprinted trait of one parent is preferably but genderaspecifically expressed in his or her offspring.

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The invention provides the initial localisation of a parentally imprinted QTL on the genome by linkage 20 analysis with genetic markers, and the actual identification of the parentally imprinted gene(s) and causal mutations therein. Molecular knowledge of such a parentally imprinted QTL allows for more efficient breeding designs herewith provided. Applications of 25 molecular knowledge of parentally imprinted QTLs in breeding programs include: marker assisted segregation analysis to identify the segregation of functionally distinct parentally imprinted QTL alleles in the populations of interest, marker assisted selection (MAS) 30 performed within lines to enhance genetic response by increasing selection accuracy, selection intensity or by reducing the generation interval using the understanding of the phenomonon of parental imprinting, marker assisted introgression (MAI) to efficiently transfer favourable 35 parentally imprinted QTL alleles from a donor to a recipient population, genetic engineering of the identified parentally QTL and genetic modification of the breeding stock using transgenic technology, development

of performance enhancing products using targeted drug development exploiting molecular knowledge of said QTL.

The inventors undertook two independent experiments to determine the practical use of parental imprinting of a QTL.

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In a first experiment, performed in a previously described Piétrain x Large White intercross, the likelihood of the data were computed under a model of paternal (paternal allele only expressed) and maternal imprinting (maternal allele only expressed) and compared 10 with the likelihood of the data under a model of a conventional "Mendelian" QTL. The results strikingly demonstrated that the QTL was indeed paternally expressed, the QTL allele (Piétrain or Large White) inherited from the F1 sow having no effect whatsoever on 15 the carcass quality and quantity of the F2 offspring. It was seen that very significant lodscores were obtained when testing for the presence of a paternally expressed QTL, while there was no evidence at all for the segregation of a QTL when studying the chromosomes 20 transmitted by the sows. The same tendency was observed for all traits showing that the same imprinted gene is responsible for the effects observed on the different traits. Table 1 reports the maximum likelihood (ML) phenotypic means for the  $F_2$  offspring sorted by inherited 25 paternal QTL allele.

In a second experiment performed in the Wild Boar X Large White intercross, QTL analyses of body composition, fatness, meat quality, and growth traits was carried out with the chromosome 2 map using a statistical model testing for the presence of an imprinting effect. Clear evidence for a paternally expressed QTL located at the very distal tip of 2p was obtained (Fig. 2; Table1). The clear paternal expression of a QTL is illustrated by the least squares means which fall into two classes following the population origin of the paternally inherited allele (Table 1). For a given paternally imprinted QTL, implementation of marker assisted segregation analysis, selection (MAS) and introgression (MAI), can be performed

using genetic markers that are linked to the QTL, genetic markers that are in linkage disequilibrium with the QTL, or using the actual causal mutations within the QTL.

Understanding the parent-of-origin effect characterising a QTL allows for its optimal use in breeding programs. Indeed, marker assisted segregation analysis under a model of parental imprinting will yield better estimates of QTL allele effects. Moreover it allows for the application of specific breeding schemes to optimally exploit a QTL. In one embodiment of the invention, the most favourable QTL alleles would be fixed in breeding animal lines and for example used to generate commercial, crossbred males by marker assisted selection (MAS, within lines) and marker assisted introgression (MAI, between lines). In another embodiment, the worst QTL alleles would be fixed in the animal lines used to generate commercial crossbred females by MAS (within lines) and MAI (between lines).

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In a preferred embodiment of the invention, said 20 animal is a pig. Note for example that the invention provides the insight that today half of the offspring from commercially popular Piétrain, Large White crossbred boars inherit an unfavourable Large White muscle mass QTL as provided by the invention causing considerable loss, and the invention now for example provides the 25 possibility to select the better half of the population in that respect. However, it is also possible to select commercial sow lines enriched with the in the boars unfavourable alleles, allowing to equip the sows with 30 other alleles more desirable for for example reproductive purposes.

In a preferred embodiment of a method provided by the invention, said QTL is located at a position corresponding to a QTL located at chromosome 2 in the pig. For example, it is known form comparative mapping data between pig and human, including bidirectional chromosome painting, that SSC2p is homologous to HSA11pter-q13<sup>11,12</sup>. HSA11pter-q13 is known to harbour a

the present invention commercially very attractive, which is even more so because the present QTL is parentally imprinted. The marker map of chromosome 2p was improved as part of this invention by adding microsatellite markers in order to cover the entire chromosome arm. The following microsatellite markers were used: Swc9, Sw2443, Sw2623, and Swr2516, all from the distal end of 2p7. QTL analyses of body composition, fatness, meat quality, and growth traits were carried out with the new chromosome 2 map. Clear evidence for a QTL located at the very distal tip of 2p was obtained (Fig. 1; Table 1). The QTL had very large effects on lean meat content in ham and explained an astonishing 30% of the residual phenotypic variance in the F2 population. Large effects on the area of the longissumus dorsi muscle, on the weight of the heart, and on back-fat thickness (subcutaneous fat) were also noted. A moderate effect on one meat quality trait, reflectance value, was indicated. The QTL had no significant effect on abdominal fat, birth weight, growth, weight of liver, kidney, or spleen (data not shown). The Large White allele at this QTL was associated with larger muscle mass and reduced back-fat thickness consistent with the difference between this breed and the Wild Boar population.

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In a second experiment, QTL mapping was performed in a Piétrain X Large White intercross comprising  $1125 \ \dot{F}_2$  offspring. The Large White and Piétrain parental breeds differ for a number of economically important phenotypes. Piétrains are famous for their exceptional muscularity and leanness  $^{10}$  (Figure 2, while Large Whites show superior growth performance. Twenty-one distinct phenotypes measuring growth performance (5), muscularity (6), fat deposition (6), and meat quality (4), were recorded on all  $F_2$  offspring. In order to map QTL underlying the genetic differences between these breeds, the inventors undertook a whole genome scan using microsatellite markers on an initial sample of 677  $F_2$  individuals. The following microsatellite marker map was used to analyse

chromosome 2;:SW2443, SWC9 and SW2623, SWR2516-(0,20)-SWR783 - (0,29) - SW240 - (0,20) - SW776 - (0,08) - S0010 - (0.04) -SW1695-(0,36)-SWR308. Analysis of pig chromosome 2 using a Maximum Likelihood multipoint algorithm, revealed highly significant lodscores (up to 20) for three of the six phenotypes measuring muscularity (% lean cuts, % ham, % loin) and three of the six phenotypes measuring fat deposition (back-fat thickness (BFT), % backfat, % fat cuts) at the distal end of the short arm of chromosome 2 (Figure 1). Positive lodscores were obtained in the 10 corresponding chromosome region for the remaining six muscularity and fatness phenotypes, however, not reaching the experiment-wise significance threshold ) ( $\alpha$ =5%. There was no evidence for an effect of the corresponding QTL on 15 growth performance (including birth weight) or recorded meat quality measurements (data not shown). To confirm this finding, the remaining sample of 355 F, offspring was genotyped for the four most distal 2p markers and QTL analysis performed for the traits yielding the highest lodscores in the first analysis. Lodscores ranged from 20 2.1 to 7.7, clearly confirming the presence of a major QTL in this region. Table 2 reports the corresponding ML estimates for the three genotypic means as well as the residual variance. Evidence based on marker assisted segregation analysis points towards residual segregation 25 at this locus within the Piétrain population.

These experiments therefore clearly indicated the existence of a QTL with major effect on carcass quality and quantity on the telomeric end of pig chromosome arm 2p; the likely existence of an allelic series at this QTL with at least three alleles: Wild-Boar < Large White < Piétrain, and possibly more given the observed segregation within the Piétrain breed.

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The effects of the identified QTL on muscle mass and
fat deposition are truly major, being of the same
magnitude of those reported for the CRC locus though
apparently without the associated deleterious effects on
meat quality. We estimate that both loci jointly explain

close to 50% of the Piétrain versus Large White breed difference for muscularity and leanness. The QTL had very large effects on lean meat content in ham and explained an astonishing 30% of the residual phenotypic variance in the  $F_2$  population. Large effects on the area of the longissumus dorsi muscle, on the weight of the heart, and on back-fat thickness (subcutaneous fat) were also noted. A moderate effect on one meat quality trait, reflectance value, was indicated. The QTL had no significant effect 10 on abdominal fat, birth weight, growth, weight of liver, kidney, or spleen (data not shown). The Large White allele at this QTL, when compared to the Wild Boar allele, was associated with larger muscle mass and reduced back-fat thickness consistent with the difference 15 between this breed and the Wild Boar population. The strong imprinting effect observed for all affected traits shows that a single causative locus is involved. The pleiotropic effects on skeletal muscle mass and the size of the heart appear adaptive from a physiological point 20 of view as a larger muscle mass requires a larger cardiac output.

In a further embodiment, the invention provides a method for selecting a pig for having desired genotypic or potential phenotypic properties comprising testing a 25 sample from said pig for the presence of a quantitative trait locus (QTL) located at a Sus scrofa chromosome 2 mapping at position 2pl.7., wherein said QTL comprises at least a part of a Sus scrofa insulin-like growth factor-2 (IGF2) allele or a genonic area closely related thereto, 30 such as polymorphisms and microsatelites and other characterising nucleic acid sequences shown herein, such as shown in figures 4 to 10. The important role of IGF2 for prenatal development is well-documented from knockout mice as well as from its causative role in the human 35 Beckwith-Wiedemann syndrome. This invention demonstrates an important role for the IGF2-region also for postnatal development.

To show the role of Igf2 the inventors performed the following three experiments:

A genomic IGF2 clone was isolated by screening a porcine BAC library. FISH analysis with this BAC clone gave a strong consistent signal on the terminal part of chromosome 2p.

A polymorphic microsatellite is located in the 3'UTR of IGF2 in mice (GenBank U71085), humans (GenBank S62623), and horse (GenBank AF020598). The possible 10 presence of a corresponding porcine microsatellite was investigated by direct sequencing of the IFG2 3'UTR using the BAC clone. A complex microsatellite was identified about 800bp downstream of the stop codon; a sequence comparison revealed that this microsatellite was identical to a previously described anonymous 15 microsatellite, Swc96. This marker was used in the initial QTL mapping experiments and its location on the genetic map correspond with the most likely position of the OTL both in the Piétrain X Large White and in the Large White 20 x Wild Boar pedigree.

Analysis of skeletal muscle and liver cDNA from 10-week old foetuses heterozygous for a nt241 (G-A) transversion in the second exon of the porcine IGFII gene and SWC9, shows that the IGFII gene is imprinted in these tissues in the pig as well and only expressed from the paternal allele.

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Based on a published porcine adult liver cDNA sequence<sup>16</sup>, the inventors designed primer pairs allowing to amplify the entire IgfII coding sequence with 222 bp of leader and 280 bp of trailor sequence from adult skeletal muscle cDNA. Piétrain and Large White RT-PCR products were sequenced indication that the coding sequences are identical in both breeds and with the published sequence. However, a G $\boxtimes$ A transition was found in the leader sequence corresponding to exon 2 in man. Following conventional nomenclature, this polymorphism will be referred to as nt241(G-A). We developed a screening test for this single nucleotide polymorphism

9(SNP) based on the ligation amplification reaction (LAR), allowing us to genotype our pedigree material. Based on these data, IgfII was shown to colocalize with the SWC9 microsatellite marker ( $\theta$ =0%), therefore virtually coinciding with the most likely position of the QTL, and well within the 95% support interval for the QTL. Subsequent sequence analysis demonstrated that the microsatellite marker SWC9 is actually located within the 3'UTR of the IgfII gene.

10 As previously mentioned, the knowledge of this QTL provides a method for the selection of animals such as pigs with improved carcass merit. Different embodiments of the invention are envisaged, including: marker assisted segregation analysis to identify the 15 segregation of functionally distinct QTL alleles in the populations of interest; marker assisted selection (MAS) performed within lines to enhance genetic response by increasing selection accuracy, selection intensity or by reducing the generation interval; marker assisted 20 introgression (MAI) to efficiently transfer favourable QTL alleles from a donor to a recipient population, thereby enhancing genetic response in the recipient population. Implementation of embodiments marker assisted segregation analysis, selection (MAS) and introgression 25 (MAI), can be performed using genetic markers that are linked to the QTL; genetic markers that are in linkage disequilibrium with the QTL, the actual causal mutations within the QTL.

In a further embodiment, the invention provides a method for selecting a pig for having desired genotypic or potential phenotypic properties comprising testing a sample from said pig for the presence of a quantitative trait locus (QTL) located at a Sus scrofa chromosome 2 mapping at position 2p1.7., wherein said QTL is paternally expressed, i.e. is expressed from the paternal allele. In man and mouse, *Igf2* is known to be imprinted and to be expressed exclusively from the paternal allele in several tissues. Analysis of skeletal muscle cDNA from

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pigs heterozygous for the SNP and/or SWC9, shows that the same imprinting holds in the pig as well. Understanding the parent-of-origin effect characterising the QTL as provided by the invention now allows for its optimal use in breeding programs. Indeed, today half of the offspring from commercially popular Piétrain x Large White crossbred boars inherit the unfavourable Large White allele causing considerable loss. Using a method as provide by the invention avoids this problem.

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The invention furthermore provides an isolated and/or recombinant nucleic acid or functional fragment derived thereof comprising a parentally imprinted quantitative trait locus (QTL) or fragment thereof capable of being predominantly expressed by one parental allele. Having such a nucleic acid as provided by the invention available allows constructing transgenic animals wherein favourable genes are capable of being exclusively or predominantly expressed by one parental allele, thereby equipping the offspring of said animal homozygous for a desired trait with desired properties related to that parental allele that is expressed.

In a preferred embodiment, the invention provides an isolated and/or recombinant nucleic acid or fragment derived thereof comprising a synthetic parentally imprinted quantitative trait locus (QTL) or functional fragment thereof derived from at least one chromosome. Synthetic herein describes a parentally expressed QTL wherein various elements are combined that originate from distinct locations from the genome of one or more animals. The invention provides recombinant nucleic acid wherein sequences related to parental imprinting of one QTL are combined with sequences relating to genes or favourable alleles of a second QTL. Such a gene construct is favourably used to obtain transgenic animals wherein the second QTL has been equipped with paternal imprinting, as opposed to the inheritance pattern in the native animal from which the second QTL is derived. Such a second QTL can for example be derived from the same

chromosome where the parental imprinting region is located, but can also be derived from a different chromosome from the same or even a different species. In the pig, such a second QTL can for example be related to an oestrogen receptor (ESR)-gene (Rothschild et al, PNAS, 93, 201-201, 1996) or a FAT-QTL (Andersson, Science, 263, 1771-1774, 1994) for example derived from an other pig chromosome, such as chromosome 4. A second or further QTL can also be derived from another (domestic) animal or a human.

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The invention furthermore provides an isolated and/or recombinant nucleic acid or functional fragment derived thereof at least partly corresponding to a QTL of a pig located at a Sus scrofa chromosome 2 mapping at position 2p1.7 wherein said QTL is related to the 15 potential muscle mass and/or fat deposition of said pig and/or wherein said QTL comprises at least a part of a Sus scrofa insulin-like growth factor-2 (IGF2) allele, preferably at least spanning a region between INS and H19, or preferably derived from a domestic pig, such as a 20 Pietrain, Meishan, Duroc, Landrace or Large White, or from a Wild Boar. For example, a genomic IGF2 clone was isolated by screening a porcine BAC library. FISH analysis with this BAC clone gave a strong consistent signal on the terminal part of chromosome 2p. A 25 polymorphic microsatellite is located in the 3'UTR of IGF2 in mice (GenBank U71085), humans (GenBank S62623), and horse (GenBank AF020598). The possible presence of a corresponding porcine microsatellite was investigated by 30 direct sequencing of the IGF2 3'UTR using the BAC clone. A complex microsatellite was identified about 800 bp downstream of the stop codon; a sequence comparison revealed that this microsatellite is identical to a previously described anonymous microsatellite, Swc9. PCR primers were designed and the microsatellite (IGF2ms) was 35 found to be highly polymorphic with three different alleles among the two Wild Boar founders and another two

among the eight Large White founders. IGF2ms was fully informative in the intercross as the breed of origin as well as the parent of origin could be determined with confidence for each allele in each  $F_2$  animal.

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A linkage analysis using the intercross pedigree was carried out with IGF2ms and the microsatellites Sw2443, Sw2623, and Swr2516, all from the distal end of  $2p^7$ . IGF2 was firmly assigned to 2p by highly significant lod scores (e.g. Z=89.0,  $\theta=0.003$  against Swr2516). Multipoint analyses, including previously typed chromosome 2 markers, revealed the following order of loci (sexaverage map distances in Kosambi cM): Sw2443/Swr2516-0.3-IGF2-14.9-Sw2623-10.3-Sw256. No recombinant was observed between Sw2443 and Swr2516, and the suggested proximal location of IGF2 in relation to these loci is based on a single recombinant giving a lod score support of 0.8 for the reported order. The most distal marker in our previous QTL study, Sw256, is located about 25 cM from the distal end of the linkage group.

20 The invention furthermore provides use of a nucleic acid or functional fragment derived thereof according to the invention in a method according to the invention. In a preferred embodiment, use of a method according to invention is provided to select a breeding animal or 25 animal destined for slaughter, or embryos or semen derived from these animals for having desired genotypic or potential phenotypic properties. In particular, the invention provides such use wherein said properties are related to muscle mass and/or fat deposition. The QTL as provided by the invention may be exploited or used to 30 improve for example lean meat content or back-fat thickness by marker assisted selection within populations or by marker assisted introgression of favorable alleles from one population to another. Examples of marker 35 assisted selection using the QTL as provided by the invention are use of marker assisted segregation analysis with linked markers or with markers in disequilibrium to identify functionally distinct QTL alleles. Furthermore, identification of a causative mutation in the QTL is now possible, again leading to identify functionally distinct QTL alleles. Such functionally distinct QTL alleles located at the distal tip of chromosome 2p with large effects on skeletal muscle mass, the size of the heart, and on back-fat thickness are also provided by the invention. The observation of a similar QTL effect in a Large White x Wild Boar as well as in a Piétrain x Large White intercross provides proof of the existence of a series of at least three distinct functional alleles. Moreover, preliminary evidence based on marker assisted segregation analysis points towards residual segregation at this locus within the Piétrain population (data not shown). The occurrence of an allelic series as provided by the invention allows identifying causal polymorphisms which - based on the quantitative nature of the observed effect - are unlikely to be gross gene alterations but rather subtle regulatory mutations. The effects on muscle mass of the three alleles rank in the same order as the breeds in which they are found i.e. Piétrain pigs are more muscular than Large White pigs that in turn have higher lean meat content than Wild Boars. The invention furthermore provides use of the alleles as provided by the invention for within line selection or for marker assisted introgression using linked markers, markers in disequilibrium or alleles comprising causative mutations.

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The invention furthermore provides an animal

selected by using a method according to the invention.

For example, a pig characterised in being homozygous for an allele in a QTL located at a Sus scrofa chromosome 2 mapping at position 2p1.7 can now be selected and is thus provided by the invention. Since said QTL is related to the potential muscle mass and/or fat deposition of said pig and/or said QTL comprises at least a part of a Sus scrofa insulin-like growth factor-2 (IGF2) allele, it is

possible to select promising pigs to be used for breeding or to be slaughtered. In particular an animal according to the invention which is a male is provided. Such a male, or its sperm or an embryo derived thereof can advantageously be used in breeding animals for creating breeding lines or for finally breeding animals destined for slaughter. In a preferred embodiment of such use as provided by the invention, a male, or its sperm, deliberately selected for being homozygous for an allele causing the extreme muscular hyperthrophy and leanness, is used to produce offspring heterozygous for such an allele. Due to said allele's paternal expression, said offspring will also show the favourable traits for example related to muscle mass, even if the parent female has a different genetic background. Moreover, it is now possible to positively select the female(s) for having different traits, for example related to fertility, without having a negative effect on the muscle mass trait that is inherited from the allele from the selected male. For example, earlier such males could occasionally be seen with Piétrain pigs but genetically it was not understood how to most profitably use these traits in breeding programmes.

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Furthermore, the invention provides a transgenic animal, sperm and an embryo derived thereof, comprising a 25 synthetic parentally imprinted QTL or functional fragment thereof as provided by the invention, i.e. it is provided by the invention to introduce a favourable recombinant allele; for example introduce the oestrogen receptor locus related to increased litter size of an animal 30 homozygously in a parentally imprinted region of a grandparent animal (for example the father of a hybrid sow if the region was paternally imprinted and the grandparent was a boar); to introduce a favourable fat-35 related allele or muscle mass-related recombinant allele in a paternally imprinted region, and so on. Recombinant alleles that are interesting or favourable from the maternal side or often the ones that have opposite effects to alleles from the paternal side. For example,

in meat animals such as pigs recombinant alleles linked with meat quality traits such as intra-muscular fat or muscle mass could be fixed in the dam lines while recombinant alleles linked with reduced back fat could be fixed in the sire lines. Other desirable combinations are for example fertility and/or milk yield in the female line with growth rates and/or muscle mass in the male lines.

The invention is further explained in the detailed description without limiting the invention.

Detailed description.

Example 1: Wild Boar x Large White intercrosses

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Methods

Isolation of an IGF2 BAC clone and fluorescent in situ hybridization (FISH). IGF2 primers (F:5'-

- GGCAAGTTCTTCCGCTAATGA-3' and R:5'-GCACCGCAGAATTACGACAA-3') for PCR amplification of a part of the last exon and 3'UTR were designed on the basis of a porcine IGF2 cDNA sequence (GenBank X56094). The primers were used to screen a porcine BAC library and the clone 253G10 was
- isolated. Crude BAC DNA was prepared as described  $^{24}$ . The BAC DNA was linearized with EcoRV and purified with QIAEXII (QIAGEN GmbH, Germany). The clone was labeled with biotin-14-dATP using the GIBCO-BRL Bionick labeling system (BRL18246-015). Porcine metaphase chromosomes were
- obtained from pokeweed (Seromed) stimulated lymphocytes using standard techniques. The slides were aged for two days at room temperature and then kept at -20 C until use. FISH analysis was carried out as previously
- described<sup>25</sup>. The final concentration of the probe in the hybridization mix was 10 ng/ $\mu$ l. Repetitive sequences were suppressed with standard concentrations of porcine

genomic DNA. After post-hybridization washing, the biotinylated probe was detected with two layers of avidin-FITC (Vector A-2011). The chromosomes were counterstained with 0.3 mg/ml DAPI (4,6-Diamino-2-phenylindole; Sigma D9542), which produced a G-banding like pattern. No posthybridization banding was needed, since chromosome 2 is easily recognized without banding. A total of 20 metaphase spreads were examined under an Olympus BX-60 fluorescence microscope connected to an IMAC-CCD S30 video camera and equipped with an ISIS 1.65 (Metasystems) software.

Sequence, microsatellite, and linkage analysis.

About two µg of linearized and purified BAC DNA was used 15 for direct sequencing with 20 pmoles of primers and BigDye Terminator chemistry (Perkin Elmer, USA). DNA sequencing was done from the 3' end of the last exon towards the 3' end of the UTR until a microsatellite was detected. A primer set (F:5'-GTTTCTCCTGTACCCACACGCATCCC-20 3' and R:5'-Fluorescein- CTACAAGCTGGGCTCAGGG-3') was designed for the amplification of the IGF2 microsatellite which is about 250 bp long and located approximately 800 bp downstream from the stop codon. The microsatellite was 25 PCR amplified using fluorescently labeled primers and the genotyping was carried out using an ABI377 sequencer and the GeneScan/Genotyper softwares (Perkin Elmer, USA). Two-point and multipoint linkage analysis were done with the Cri-Map software<sup>26</sup>.

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Animals and phenotypic data.

The intercross pedigree comprised two European Wild Boar males and eight Large White females, 4  $F_1$  males and 22  $F_1$  females, and 200  $F_2$  progeny<sup>1</sup>. The  $F_2$  animals were sacrificed at a live weight of at least 80 kg or at a

maximum age of 190 days. Phenotypic data on birth weight, growth, fat deposition, body composition, weight of internal organs, and meat quality were collected; a detailed description of the phenotypic traits are provided by Andersson  $et\ al.^{1}$  and Andersson-Eklund  $et\ al.^{4}$ 

Statistical analysis.

Interval mapping for the presence of QTL were carried out 10 with a least squares method developed for the analysis of crosses between outbred lines $^{27}$ . The method is based on the assumption that the two divergent lines are fixed for alternative QTL alleles. There are four possible 15 genotypes in the  $F_2$  generation as regards the grandparental origin of the alleles at each locus. This makes it possible to fit three effects: additive, dominance, and imprinting $^2$ . The latter is estimated as the difference between the two types of heterozygotes, the one receiving the Wild Boar allele through an  $F_1$  sire 20 and the one receiving it from an  $F_1$  dam. An F-ratio was calculated using this model (with 3 d.f.) versus a reduced model without a OTL effect for each cM of chromosome 2. The most likely position of a QTL was 25 obtained as the location giving the highest F-ratio. Genome-wise significance thresholds were obtained empirically by a permutation test $^{28}$  as described $^{2}$ . The QTL model including an imprinting effect was compared with a model without imprinting (with 1 d.f.) to test 30 whether the imprinting effect was significant.

The statistical models also included the fixed effects and covariates that were relevant for the respective traits; see Andersson-Eklund  $et\ al.\ ^4$  for a more detailed description of the statistical models used.

35 Family was included to account for background genetic

effects and maternal effects. Carcass weight was included as a covariate to discern QTL effects on correlated traits, which means that all results concerning body composition were compared at equal weights. Least-squares means for each genotype class at the IGF2 locus were 5 estimated with a single point analysis using Procedure GLM of  $SAS^{29}$ ; the model included the same fixed effects and covariates as used in the interval mapping analyses. The QTL shows a clear parent of origin-specific expression and the map position coincides with that of 10 the insulin-like growth factor II gene (IGF2), indicating IGF2 as the causative gene. A highly significant segregation distortion (excess of Wild Boar-derived alleles) was also observed at this locus. The results 15 demonstrate an important effect of the IGF2 region on postnatal development and it is possible that the presence of a paternally expressed IGF2-linked QTL in humans and in rodent model organisms has so far been overlooked due to experimental design or statistical treatment of data. The study has also important 20 implications for quantitative genetics theory and practical pig breeding.

IGF2 was identified as a positional candidate gene for this QTL due to the observed similarity between pig chromosome 2p and human chromosome 11p. A genomic IGF2 25 clone was isolated by screening a porcine BAC library. FISH analysis with this BAC clone gave a strong consistent signal on the terminal part of chromosome 2p (Fig. 1). A polymorphic microsatellite is located in the 30 3'UTR of IGF2 in mice (GenBank U71085), humans (GenBank S62623), and horse (GenBank AF020598). The possible presence of a corresponding porcine microsatellite was investigated by direct sequencing of the IGF2 3'UTR using the BAC clone. A complex microsatellite was identified 35 about 800 bp downstream of the stop codon; a sequence comparison revealed that this microsatellite is identical to a previously described anonymous microsatellite,  $Swc9^6$ . PCR primers were designed and the microsatellite (IGF2ms) was found to be highly polymorphic with three different alleles among the two Wild Boar founders and another two among the eight Large White founders. IGF2ms was fully informative in the intercross as the breed of origin as well as the parent of origin could be determined with confidence for each allele in each  $F_2$  animal.

10 A linkage analysis using the intercross pedigree was carried out with IGF2ms and the microsatellites Sw2443, Sw2623, and Swr2516, all from the distal end of  $2p^7$ . IGF2was firmly assigned to 2p by highly significant lod scores (e.g. Z=89.0,  $\theta$ =0.003 against Swr2516). Multipoint analyses, including previously typed chromosome 2 15  $markers^8$ , revealed the following order of loci (sexaverage map distances in Kosambi cM): Sw2443/Swr2516-0.3-IGF2-14.9-Sw2623-10.3-Sw256. No recombinant was observed between Sw2443 and Swr2516, and the suggested proximal location of IGF2 in relation to these loci is based on a 20 single recombinant giving a lod score support of 0.8 for the reported order. The most distal marker in our previous QTL study, Sw256, is located about 25 cM from the distal end of the linkage group.

QTL analyses of body composition, fatness, meat quality, and growth traits were carried out with the new chromosome 2 map using a statistical model testing for the possible presence of an imprinting effect as expected for IGF2. Clear evidence for a paternally expressed QTL located at the very distal tip of 2p was obtained (Fig. 2; Table 1). The QTL had very large effects on lean meat content in ham and explained an astonishing 30% of the residual phenotypic variance in the F2 population. Large effects on the area of the longissumus dorsi muscle, on the weight of the heart, and on back-fat thickness

(subcutaneous fat) were also noted. A moderate effect on one meat quality trait, reflectance value, was indicated. The QTL had no significant effect on abdominal fat, birth weight, growth, weight of liver, kidney, or spleen (data not shown). The Large White allele at this QTL was associated with larger muscle mass and reduced back-fat thickness consistent with the difference between this breed and the Wild Boar population. The strong imprinting effect observed for all affected traits strongly suggests a single causative locus. The pleiotropic effects on skeletal muscle mass and the size of the heart appear adaptive from a physiological point of view as a larger muscle mass requires a larger cardiac output. The clear paternal expression of this QTL is illustrated by the least squares means which fall into two classes following the population origin of the paternally inherited allele (Table 1). It is worth noticing though that there was a non-significant trend towards less extreme values for the two heterozygous classes, in particular for the estimated effect on the area of longissimus dorsi. This may be due to chance, but could have a biological explanation, e.g. that there is some expression of the maternally inherited allele or that there is a linked, non-imprinted QTL with minor effects on the traits in question.

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The IGF2-linked QTL and the FAT1 QTL on chromosome 4

1, 9 are by far the two loci with the largest effect on body composition and fatness segregating in this Wild Boar intercross. The IGF2 QTL controls primarily muscle mass whereas FAT1 has major effects on fat deposition

30 including abdominal fat, a trait that was not affected by the IGF2 QTL (Fig. 2). No significant interaction between the two loci was indicated and they control a very large proportion of the residual phenotypic variance in the F2 generation. A model including both QTLs explains 33.1% of the variance for percentage lean meat in ham, 31.3% for the percentage of lean meat plus bone in back, and 26.2%

for average back fat depth (compare with a model including only chromosome 2 effects, Table 1). The two QTLs must have played a major role in the response during selection for lean growth and muscle mass in the Large White domestic pig.

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A highly significant segregation distortion was observed in the IGF2 region (excess of Wild Boar-derived alleles) as shown in Table 1 ( $\chi$ 2=11.7, d.f.=2; P=0.003). The frequency of Wild Boar-derived IGF2 alleles was 59% in contrast to the expected 50% and there was twice as many "Wild Boar" as "Large White" homozygotes. This deviation was observed with all three loci at the distal tip and is thus not due to typing errors. The effect was also observed with other loci but the degree of distortion decreased as a function of the distance to the distal tip of the chromosome. Blood samples for DNA preparation were collected at 12 weeks of age and we are convinced that the deviation from expected Mendelian ratios was present at birth as the number of animals lost prior to blood sampling was not sufficient to cause a deviation of this magnitude. No other of the more than 250 loci analyzed in this pedigree show such a marked segregation distortion (L. Andersson, unpublished). The segregation distortion did not show an imprinting effect, as the frequencies of the two reciprocal types of heterozygotes were identical (Table 1). This does not exclude the possibility that the QTL effects and the segregation distortion are controlled by the same locus. The segregation distortion maybe due to meiotic drive favoring the paternally expressed allele during gametogenesis, as the F<sub>1</sub> parents were all sired by Wild Boar males. Another possibility is that the segregation distortion may be due to codominant expression of the maternal and paternal allele in some tissues and/or during a critical period of embryo development. Biallelic IGF2 expression has been reported to occur to some extent

during human development $^{10}$ ,  $^{11}$  and interestingly a strong influence of the parental species background on IGF2 expression was recently found in a cross between Mus musculus and Mus spretus12. It is also interesting that a VNTR polymorphism at the insulin gene, which is very closely linked to IGF2, is associated with size at birth in humans  $^{13}$ . It is possible that the IGF2-linked QTL in pigs has a minor effect on birth weight but in our data it was far from significant (Fig. 2) and there was no indication of an imprinting effect.

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This study is an advance in the general knowledge concerning the biological importance of the IGF2 locus. The important role of IGF2 for prenatal development is well-documented from knock-out mice $^{14}$  as well as from its causative role in the human Beckwith-Wiedemann  $syndrome^{15}$ . This study demonstrates an important role for the IGF2-region also for postnatal development. It should be stressed that our intercross between outbred populations is particularly powerful to detect QTL with a parent of origin-specific effect on a multifactorial trait. This is because multiple alleles (or haplotypes) are segregating and we could deduce whether a heterozygous F2 animal received the Wild Boar allele from the  $F_1$  male or female. It is quite possible that the segregation of a paternally expressed IGF2-linked QTL affecting a trait like obesity has been overlooked in human studies or in intercrosses between inbred rodent populations because of experimental design or statistical treatment of data. An imprinting effect cannot be detected in an intercross between two inbred lines as only two alleles are segregating at each locus. Our result has therefore significant bearings on the future analysis of the association between genetic polymorphism in the insulin-IGF2 region and Type I diabetes 16, obesity 17, and variation in birth weight 13 in humans, as 35

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well as for the genetic dissection of complex traits using inbred rodent models. A major impetus for generating an intercross between the domestic pig and its wild ancestor was to explore the possibilities to map and identify major loci that have responded to selection. We have now showed that two single QTLs on chromosome 2 (this study) and  $4^{1}$ , 2 explain as much as one third of the phenotypic variance for lean meat content in the F2 generation. This is a gross deviation from the underlying assumption in the classical infinitesimal model in quantitative genetics theory namely that quantitative traits are controlled by an infinite number of loci each with an infinitesimal effect. If a large proportion of the genetic difference between two divergent populations (e.g. Wild Boar and Large White) is controlled by a few loci, one would assume that selection would quickly fix OTL alleles with large effects leading to a selection plateau. However, this is not the experience in animal breeding programs or selection experiments where good persistent long-term selection responses are generally obtained, provided that the effective population size is reasonably large 18. A possible explanation for this paradox is that QTL alleles controlling a large proportion of genetic differences between two populations may be due to several consecutive mutations; this may be mutations in the same gene or at several closely linked genes affecting the same trait. It has been argued that new mutations contribute substantially to long-term selection responses 19, but the genomic distribution of such mutations are unknown.

The search for a single causative mutation is the paradigm as regards the analysis of genetic defects in mice and monogenic disorders in humans. We propose that this may not be the case for loci that have been under selection for a large number of generations in domestic animals, crops, or natural populations. This hypothesis

predicts the presence of multiple alleles at major QTL. It gains some support from our recent characterization of porcine coat color variation. We have found that both the alleles for dominant white color and for black-spotting differ from the corresponding wild-type alleles by at least two consecutive mutations with phenotypic effects at the KIT and MCIR loci, respectively<sup>20</sup>, 21. In this context it is highly interesting that in the accompanying example we have identified a third allele at the IGF2-linked QTL. The effects on muscle mass of the three alleles rank in the same order as the breeds in which they are found i.e. Piétrain pigs are more muscular than Large White pigs that in turn have higher lean meat content than Wild Boars.

There are good reasons to decide that IGF2 is the 15 causative gene for the now reported OTL. Firstly, there is a perfect agreement in map localization (Fig. 2). Secondly, it has been shown that IGF2 is paternally expressed in mice, humans, and now in pigs, like the QTL. There are several other imprinted genes in the near 20 vicinity of IGF2 in mice and humans (Mash2, INS2, H19, KVLQT1, TAPA1/CD81, and CDKN1C/p57KIP2) but only IGF2 is paternally expressed in adult tissues<sup>22</sup>. We believe that this locus provides a unique opportunity for molecular characterization of a QTL. The clear paternal expression 25 can be used to exclude genes that do not show this mode of inheritance. Moreover, the presence of an allelic series should facilitate the difficult distinction between causative mutations and linked neutral polymorphism. We have already shown that there is no 30 difference in coding sequence between IGF2 alleles from Piétrain and Large White pigs suggesting that the causative mutations occur in regulatory sequences. An obvious step is to sequence the entire IGF2 gene and its multiple promoters from the three populations. The recent 35

report that a VNTR polymorphism in the promoter region of the insulin (INS) gene affects IGF2 expression<sup>23</sup> suggests that the causative mutations may be at a considerable distance from the IGF2 coding sequence.

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The results have several important implications for the pig breeding industry. They show that genetic imprinting is not an esoteric academic question but need to be considered in practical breeding programs. The detection of three different alleles in Wild Boar, Large White, and Piétrain populations indicates that further alleles at the IGF2-linked QTL segregate within commercial populations. The paternal expression of the QTL facilitates its detection using large paternal half-sib families as the female contribution can be ignored. The QTL is exploited to improve lean meat content by marker assisted selection within populations or by marker assisted introgression of favorable alleles from one population to another.

Example 2: Piétrain x Large White intercrosses

### Methods

Pedigree material: The pedigree material utilized to map

QTL was selected from a previously described Piétrain x

Large White F2 pedigree comprising > 1,800 individuals<sup>6,7</sup>.

To assemble this F2 material, 27 Piétrain boars were

mated to 20 Large White sows to generate an F1 generation

comprising 456 individuals. 31 F1 boars were mated to

unrelated 82 F1 sows from 1984 to 1989, yielding a total

of 1862 F2 offspring. F1 boars were mated on average to 7

females, and F1 sows to an average of 2,7 males. Average

offspring per boar were 60 and per sow 23.

- 15 Phenotypic information: (i) Data collection: A total of 21 distinct phenotypes were recorded in the F2 generation<sup>6,7</sup>. These included:
  - five growth traits: birth weight (g), weaning weight (Kg), grower weight (Kg), finisher weight (Kg) and
- 20 average daily gain (ADG; Kg/day; grower to finsher period);
  - two body proportion measurements: carcass length (cm);
    and a conformation score (0 to 10 scale; ref.6);
  - ten measurements of carcass composition obtained by
- dissection of the chilled carcasses 24 hours after slaughter. These include measurements of muscularity: % ham (weight hams/carcass weight), % loin (weight loin/carcass weight), % shoulder ( weight
- shoulder/carcass weight), % lean cuts (% ham + %loin + %
- shoulder); and measurements of fatness: average back-fat thickness (BFT; cm), % backfat (weight backfat/carcass weight), % belly (weight belly/carcass weight), % leaf fat (weight leaf fat/carcass weight), % jowl (weight jowl/carcass weight), and "% fat cuts" (% backfat + %
- 35 belly + % leaft fat + % jowl).
  - four meat quality measurements: pH  $_{ exttt{LD1}}$  (Longissimus dorsi 1

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hour after slaughter), pH  $_{\text{LD24}}$  (Longissimus dorsi 24 hours after slaughter), pH  $_{\text{G1}}$  (Gracilis 1 hour after slaughter) and pH  $_{\text{G24}}$  (Gracilis 24 hours after slaughter). (ii) Data processing: Individual phenotypes were preadjusted for fixed effects (sire, dam, CRC genotype, sex, year-season, parity) and covariates (litter size, birth weight, weaning weight, grower weight, finisher weight) that proved to significantly affect the corresponding trait. Variables included in the model were selected by stepwise regression.

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Marker genotyping: Primer pairs utilized for PCR amplification of microsatellite markers are as described<sup>19</sup>. Marker genotyping was performed as previously described<sup>20</sup>. Genotypes at the CRC and MyoD loci were determined using conventional methods as described<sup>1,12</sup>. The LAR test for the Igf2 SNP was developed according to Baron et al.<sup>21</sup> using a primer pair for PCR amplification (5'-CCCCTGAACTTGAGGACGAGCCGC-3';5'-ATCGCTGTGGGCTGGGCTGGCC-3') and a set of three primers for the LAR step (5'-FAM-CGCCCCAGCTGCCCCCCAG-3'; 5'-HEX-CGCCCCAGCTGCCCCCAA-3'; 5'-CCTGAGCTGCAGCAGGCCAG-3').

Map construction: Marker maps were constructed using the TWOPOINT, BUILD and CHROMPIC options of the CRIMAP package<sup>22</sup>. To allow utilisation of this package, full-sib families related via the boar or sow were disconnected and treated independently. By doing so, some potentially usable information was neglected, yielding, however, unbiased estimates of recombination rates.

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QTL mapping: (i) Mapping Mendelian QTL: Conventional QTL mapping was performed using a multipoint maximum likelihood method. The applied model assumed one segregating QTL per

chromosome, and fixation of alternate QTL alleles in the respective parental lines, Piétrain (P) and Large White (LW). A specific analysis program had to be developed to account for the missing genotypes of the parental generation, resulting in the fact that the parental origin of the F1 chromosomes could not be determined. Using a typical "interval mapping" strategy, an hypothetical QTL was moved along the marker map using user-defined steps. At each position, the likelihood (L) of the pedigree data was computed as:

$$L = \sum_{\varphi=1}^{2'} \prod_{i=1}^{n} \sum_{G=1}^{4} (P(G|M_i, \theta, \varphi)P(y_i|G))$$

P or right chromosme P), there is a total of  $2^r$  combinations for r F1 parents.

$$15 \quad \prod_{i=1}^{n} \quad n \quad F2$$

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 $\sum_{G=1}^{4}$  ith F2 offspring, over the four possible QTL genotypes:

P/P, P/LW, LW/P and LW/LW

 $P(G|M_i,\theta,\phi)M_i$ : the marker genotype of the *i*th F2 offspring and its F1 parents, (ii): the vector of recombination rates between adjacent markers and between the hypothetical QTL and its flanking markers, and (iii) $\theta$  the considered marker-QTL phase combination of the F1 parents.

Recombination rates and marker linkage phase of Fl parents are assumed to be known when computing this probability. Both were determined using CRIMAP in the map construction phase (see above).

 $P(y_i|G)\,y_i)$  of offspring i, given the QTL genotype under consideration. This probability is computed from the normal density function:

$$P(y_i|G) = \frac{1}{\sqrt{2\pi\sigma}}e^{\frac{-(y_i-\mu_G)^2}{2\sigma^2}}$$

 $_{G}$  is the phenotypic mean of the considered QTL genotype (PP, PL, LP or LL) and  $\sigma^{2}$  the residual variance  $\sigma^{2}$  was considered to be the same for the four QTL genotypic classes.

5 The values of  $\mu_{PP}$ ,  $\mu_{PL}=\mu_{LP}$ ,  $\mu_{LL}$  and  $\sigma^2$  maximizing L were determined using the GEMINI optimisation routine<sup>23</sup>.

The likelihood obtained under this alternative  $H_1$  hypothesis was compared with the likelihood obtained under the null hypothesis  $H_0$  of no QTL, in which the phenotypic means of the

10 four QTL genotypic classes were forced to be identical. The difference between the logarithms of the corresponding likelihoods yields a lodscore measuring the evidence in favour of a QTL at the corresponding map position.

(ii) Significance thresholds: Following Lander & Botstein 24,

lodscore thresholds (T) associated with a chosen genome-wise significance level, were computed such that:

$$\alpha = (C + 9.21GT)\chi_2^2(4.6T)$$

 ${\it C}$  corresponds to the number of chromosomes (= 19),  ${\it G}$  corresponds to the length of the genome in Morgans (= 29),

and  $\chi^2_1$  (4.6T) denotes one minus the cumulative distribution function of the chi-squared distribution with 2 d.f. Single point  $2\ln(LR)$  were assumed to be distributed as a chi-squared distribution with two degrees of freedom, as we were fitting both an additive and dominance component. To account for the

fact that we were analysing multiple traits, significance levels were adjusted by applying a Bonferoni correction corresponding to the effective number of independent traits that were analyzed. This effective number was estimated at 16 following the approach described by Spelman et al.<sup>25</sup>.

30 Altogether, this allowed us to set the lodscore threshold associated with an experiment-wise significance level of 5%

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at 5.8. When attempting to confirm the identified QTL in an independent sample, the same approach was used, however, setting C at 1, G at 25cM and correcting for the analysis of 4.5 independent traits (as only six traits were analyzed in this sample). This yielded a lodscore threshold associated with a Type I error of 5% of 2.

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(iii). Testing for an imprinted QTL: To test for an imprinted QTL, we assumed that only the QTL alleles transmitted by the parent of a given sex would have an effect on phenotype, the QTL alleles transmitted by the other parent being "neutral". The likelihood of the pedigree data under this hypothesis was computed using equation 1. To compute  $P(y_i \mid G)$ , however, the phenotypic means of the four QTL genotypes were set at  $\mu_{PP}$  =  $\mu_{PL} = \mu_{P}$  and  $\mu_{LP} = \mu_{LL} = \mu_{L}$  to test for a QTL for which the paternal allele only is expressed, and  $\mu_{PP}$  =  $\mu_{LP}$  =  $\mu_P$  and  $\mu_{PL}$  =  $\mu_{LL} = \mu_{L}$  to test for a QTL for which the maternal allele only is expressed. It is assumed in this notation that the first subscript refers to the paternal allele, the second subscript to the maternal allele.  $H_0$  was defined as the null-hypothesis of no QTL,  $H_1$  testing the presence of a Mendelian QTL;  $H_2$ testing the presence of a paternally expressed QTL, and  $H_3$ testing the presence of a maternally expressed QTL.

RT-PCR: Total RNA was extracted from skeletal muscle

according to Chirgwin et al. 26. RT-PCR was performed using
the Gene-Amp RNA PCR Kit (Perkin-Elmer) The PCR products were
purified using QiaQuick PCR Purification kit (Qiagen) and
sequenced using Dye terminator Cycle Sequencing Ready
Reaction (Perkin Elmer) and an ABI373 automatic sequencer.

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In example 2 we report the identification of a QTL with major effect on muscle mass and fat deposition mapping to porcine 2p1.7 The QTL shows clear evidence for parental imprinting strongly suggesting the involvement of the *Igf2* locus.

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A Piétrain X Large White intercross comprising 1125  $F_2$  offspring was generated as described<sup>6,7</sup>. The Large White and Piétrain parental breeds differ for a number of economically important phenotypes. Piétrains are famed for their exceptional muscularity and leannes <sup>8</sup> (Figure 2), while Large Whites show superior growth performance. Twenty-one distinct phenotypes measuring (i) growth performance (5), (ii) muscularity (6), (iii) fat deposition (6), and (iv) meat quality (4), were recorded on all  $F_2$  offspring.

In order to map QTL underlying the genetic differences between these breeds, we undertook a whole genome scan using microsatellite markers on an initial sample of  $677 ext{ } ext{F}_2$ individuals. Analysis of pig chromosome 2 using a ML multipoint algorithm, revealed highly significant lodscores (up to 20) for six of the 12 phenotypes measuring muscularity and fat deposition at the distal end of the short arm of chromosome 2 (Figure 3a). Positive lodscores were obtained for the remaining six phenotypes, however, not reaching the genome-wise significance threshold ( = 5%). To confirm this finding, the remaining sample of 355  $F_2$  offspring was genotyped for the five most distal 2p markers and QTL analysis performed for the traits yielding the highest lodscores in the first analysis. Lodscores ranged from 2.1 to 7.7, clearly confirming the presence of a major QTL in this region. Table 2 reports the corresponding ML estimates for the three genotypic means as well as the corresponding residual variance.

Bidirectional chromosome painting establishes a correspondence between SSC2p and HSA11pter-q13<sup>9,10</sup>. At least

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two serious candidate genes map to this region in man: the myogenic basic helix-loop-helix factor, MyoD, maps to HSA11p15.4, while Igf2 maps to HSA11p15.5 MyoD is a well known key regulator of myogenesis and is one of the first myogenic markers to be switched on during development 11. A 5 previously described amplified sequence polymorphism in the porcine MyoD gene12 proved to segregate in our F2 material, which was entirely genotyped for this marker. analysis positioned the MyoD gene in the SW240-SW776 (odds > 1000) interval, therefore well outside the lod-2 drop off 10 support interval for the QTL (figure 1). Igf2 is known to enhance both proliferation and differentiation of myoblasts in vitro<sup>13</sup> and to cause a muscular hypertrophy when overexpressed in vivo. Based on a published porcine adult liver cDNA sequence<sup>14</sup>, we designed primer pairs allowing us 15 to amplify the entire Iqf2 coding sequence with 222 bp of leader and 280 bp of trailor sequence from adult skeletal muscle cDNA. Piétrain and Large White RT-PCR products were sequenced indicating that the coding sequences was identical in both breeds and with the published sequence. However, a G 20 A transition was found in the leader sequence corresponding to exon 2 in man (Figure 4). We developed a screening test for this single nucleotide polymorphism (SNP) based on the ligation amplification reaction (LAR), allowing us to genotype our pedigree material. Based on these data, Igf2 was 25 shown to colocalize with the SWC9 microsatellite marker ( = 0%), therefore located at approximately 2 centimorgan from the most likely position of the QTL and well within the 95% support interval for the QTL (figure 1). Subsequent sequence analysis demonstrated that the microsatellite marker SWC9 is 30 actually located within the 3' UTR of the Igf2 gene. Combined with available comparative mapping data for the PGA and FSH loci, these results suggest the occurrence of an interstitial

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inversion of a chromosome segment containing MyoD, but not Igf2 which has remained telomeric in both species.

Igf2 therefore appeared as a strong positional allele having the observed QTL effect. In man and mouse, Iqf2 is known to be imprinted and to be expressed exclusively from the paternal allele in several tissues15. Analysis of skeletal muscle cDNA from pigs heterozygous for the SNP and/or SWC9, shows that the same imprinting holds in this tissue in the pig as well (Figure 4). Therefore if Iqf2 were responsible for the observed effect, and knowing that only the paternal Igf2 allele is expressed, one can predict that (i) the paternal allele transmitted by F1 boars (P or LW) would have an effect on phenotype of F2 offspring, (ii) the maternal allele transmitted by F1 sows (P or LW) would have no effect on phenotype of F2 offspring, and (iii) the likelihood of the data would be superior under a model of a bimodal (1:1) F2 population sorted by inherited paternal allele when compared to a conventional "Mendelian" model of a trimodal (1:2:1) F2 population. The QTL mapping programs were adapted in order to allow testing of the corresponding hypotheses. Ho was defined as the null-hypothesis of no QTL,  $H_1$  as testing for the presence of a Mendelian QTL,  $H_2$  as testing for the presence of a paternally expressed QTL, and  ${\rm H}_{3}$  as testing for the presence of a maternally expressed QTL.

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Figure 3 summarizes the obtained results. Figure 3a, 3b and 3c respectively show the lodscore curves corresponding to  $\log_{10}~(H_2/H_0)$ ,  $\log_{10}~(H_3/H_0)$  and  $\log_{10}~(H_2/H_1)$ . It can be seen that very significant lodscores are obtained when testing for the presence of a paternally expressed QTL, while there is no evidence at all for the segregation of a QTL when studying the chromosomes transmitted by the sows. Also, the hypothesis of a paternally expressed QTL is significantly more likely (  $\log_{10}~(H_2/H_1)~>~3$ ) than the hypothesis of a "Mendelian" QTL

for all examined traits. The fact that the same tendency is observed for all traits indicates that it is likely the same imprinted gene that is responsible for the effects observed on the different traits. Table 2 reports the ML phenotypic means for the F2 offspring sorted by inherited paternal QTL allele. Note that when performing the analysis under a model of a mendelian QTL, the Piétrain and Large White QTL alleles appeared to behave in an additive fashion, the heterozygous genotype exhibiting a phenotypic mean corresponding exactly to the midpoint between the two homzygous genotypes. This is exactly what one would predict when dealing with an imprinted QTL as halve of the heterozygous offspring are expected to have inherited the P allele from their sire, the other halve the LW allele.

These data therefore confirmed our hypothesis of the 15 involvement of an imprinted gene expressed exclusively from the paternal allele. The fact that the identified chromosomal segment coincides precisely with an imprinted domain documented in man and mice strongly implicates the 20 orthologous region in pigs. At least seven imprinted genes mapping to this domain have been documented (Igf2, Ins2, H19, Mash2,  $p57^{\text{KIP2}}$ ,  $K_{\text{v}}LQTL1$  and TDAG51) (ref. 15 and Andrew Feinberg, personal communication). Amongst these, only Igf2and Ins2 are paternally expressed. While we cannot exclude that the observed QTL effect is due to an as of yet 25 unidentified imprinted gene in this region, its reported effects on myogenesis in vitro and in vivo 13 strongly implicate Igf2. Particularly the muscular hypertrophy observed in transgenic mice overexpressing Igf2 from a muscle specific promotor are in support of this hypothesis (Nadia 30 Rosenthal, personal communication. Note that allelic variants of the  ${\it INS}$  VNTR have recently been shown to be associated

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with size at birth in  $man^{16}$ , and that the same VNTR has been shown to affect the level of Igf2 expression<sup>17</sup>.

The observation of the same QTL effect in a Large White x Wild Boar intercross indicates the existence of a series of at least three distinct functional alleles. Moreover, preliminary evidence based on marker assisted segregation analysis points towards residual segregation at this locus within the Piétrain population (data not shown). The occurrence of an allelic series might be invaluable in identifying the causal polymorphisms which - based on the quantitatve nature of the observed effect - are unlikely to be gross gene alterations but rather subtle regulatory mutations.

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The effects of the identified QTL on muscle mass and fat deposition are truly major, being of the same magnitude of those reported for the CRC locus<sup>6,7</sup> though apparently without the associated deleterious effects on meat quality. We estimate that both loci jointly explain close to 50% of the Piétrain versus Large White breed difference for muscularity and leanness. Understanding the parent-of-origin effect characterizing this locus will allow for its optimal use in breeding programs. Indeed, today half of the offspring from commercially popular Piétrain x Large White crossbred boars inherit the unfavourable Large White allele causing considerable loss.

The QTL described in this work is the second example of a gene affecting muscle development in livestock species that exhibits a non-mendelian inheritance pattern. Indeed, we have previously shown that the callipyge locus (related to the qualitative trait wherein muscles are doubled) is characterized by polar overdominance in which only the heterozygous individuals that inherit the CLPG mutation from their sire express the double-muscling phenotype<sup>5</sup>. This

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demonstrates that parent-of-origin effects affecting genes underlying production traits in livestock might be relatively common.

# 5 Example 3:

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Generating a reference sequence of IGF2 and flanking loci in the pig.

The invention provides an imprinted QTL with major effect on muscle mass mapping to the IGF2 locus in the pig, and use of the QTL as tool in marker assisted selection. To fine tune this tool for marker assisted selection, as well as to further identify a causal mutation, we have further generated a reference sequence encompassing the entire porcine IGF2 sequence as well as that from flanking genes.

To achieve this, we screened a porcine BAC library with IGF2 probes and identified two BACs. BAC-PIGF2-1 proved to contain the INS and IGF2 genes, while BAC-PIGF2-2 proved to contain the IGF2 and H19 genes. The NotI map as well as the relative position of the two BACs is shown in Figure 5. BAC-PIGF2-1 was shotgun sequenced using standard procedures and automatic sequencers. The resulting sequences were assembled using standard software yielding a total of 115 contigs. The corresponding sequences are reported in figure 6. Similarity searches were performed between the porcine contigs and the orthologous sequences in human. Significant homologies were detected for 18 contigs and are reported in Figure 7.

For BAC-PIGF2-2, the 24 Kb NotI fragment not present in BAC-PIGF2-1 was subcloned and sequenced using the EZ::TN transposon approach and ABI automatic sequencers. Resulting

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sequences were assembled using the Phred-Phrap-Consed program suit, yielding seven distinct contigs (figure 8). The contig sequences were aligned with the corresponding orthologous human sequences using the compare and dotplot programs of the GCG suite. Figure 9 symmarizes the corresponding results.

Example 4: Identification of DNA sequence polymorphisms in the IGF2 and flanking loci.

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Based on the reference sequence obtained as described in Example 1, we resequenced part of the IGF2 and flanking loci from genomic DNA isolated from Piétrain, Large White and Wild Boar individuals, allowing identification of DNA sequence polymorphisms such as reported in figure 10.

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Legends to the figures

Fig. 1: Test statistic curves obtained in QTL analyses of

5 chromosome 2 in a Wild Boar/Large White intercross. The graph
plots the F ratio testing the hypothesis of a single QTL at a
given position along the chromosome for the traits indicated.
The marker map with the distances between markers in Kosambi
centiMorgan is given on the X-axis. The horizontal lines

10 represent genome-wise significant (P<0.05) and suggestive
levels for the trait lean meat in ham; similar significance
thresholds were obtained for the other traits.

Figure 2: Piétrain pig with characteristic muscular hypertrophy.

Figure 3: Lodscore curves obtained in a Piétrain x Large White intercross for six phenotypes measuring muscle mass and fat deposition on pig chromosome 2. The most likely positions of the Igf2 and MyoD genes determined by linkage analysis with respect to the microsatellite marker map are shown.  $H_0$  was defined as the null-hypothesis of no QTL,  $H_1$  as testing for the presence of a Mendelian QTL,  $H_2$  as testing for the presence of a paternally expressed QTL, and  $H_3$  as testing for the presence of a maternally expressed QTL.  $H_1$  3a:  $H_2$  1b:  $H_3$  1c:  $H_3$ 

Figure 4: A. Structure of the human *Igf2* gene according to ref. 17, with aligned porcine adult liver cDNA sequence as reported in ref. 16. The position of the *nt241(G-A)* transition and *Swc9* microsatellite are shown. B. The corresponding markers were used to demonstrate the monoallelic (paternal) expression of *Igf2* in skeletal muscle

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and liver of 10-week old fetuses. PCR amplification of the  $nt421\,(G-A)$  polymorphism and Swc9 microsatellite from genomic DNA clearly shows the heterozygosity of the fetus, while only the paternal allele is detected in liver cDNA  $(nt421\,(G-A))$  and Swc9) and muscle cDNA (Swc9). The absence of RT-PCR product for  $nt421\,(G-A)$  from in fetal muscle points towards the absence of mRNA including exon 2 in this tissue. Parental origin of the foetal alleles was determined from the genotypes of sire and dam (data not shown).

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Figure 5: A NotI restriction map showing the relative position of BAC-PIGF2-1 (comprising INS and IGF2 genes), and BAC-PIGF2-2 (comprising IGF2 and H19 genes).

15 Figure 6: Nucleic acid sequences of contig 1 to contig 115 derived from BAC-PIGF2-1 which was shotgun sequenced using standard procedures and automatic sequencers.

Figure 7: Similarity between porcine contigs of figure 6 and orthologous sequences in human.

Figure 8 Nucleic acid sequences of contig 1 to contig 7 derived from BAC-PIGF2-2, (the 24 Kb NotI fragment not present in BAC-PIGF2-1) which was subcloned and sequenced using the EZ::TN transposon approach and ABI automatic sequencers.

Figure 9: Similarity between porcine contigs of figure 8 and orthologous sequences in human.

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Figure 10: DNA sequence polymorphisms in the IGF2 and flanking loci from genomic DNA isolated from Piétrain, Large White and Wild Boar individuals.

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	Table 1 Summary of OTI, analysis for nig chromosome 2 in a Wild Boar/Large White intercross <sup>1</sup>	sis for pig chr	omosome 2 in	a Wild Boar/	Large White intercre	JSS <sup>1</sup>			vo I
	Trait	F ratio <sup>2</sup>	Imprinting	Map position <sup>3</sup>	Percent of F <sub>2</sub>	Least squares means <sup>5</sup> WP/WM WP/LM	es means <sup>5</sup> WP/LM	LP/WM	00/3614
	$\Gamma_{P/LM}$	i N	0						13
ß						n=62	n=43	n=43	n=30
	Body composition traits								
	Lean meat in ham, %	24.4**	19.1***	0	30.6	63.6	64.2	.66.4 <sup>b</sup>	67.3 <sup>b</sup>
	Lean meat mass in ham, kg	18.1***	16.8***		24.3	4.69	4.72	4.94 <sup>b</sup>	5.02 <sup>b</sup>
10	Lean meat + bone in back, %	12.2**	**9.6	0	17.4	66.3	.2.99	69.3°	70.8°
	Longissimus muscle area, cm²	10.3**	4.8*	-	15.4	31.9	33.0	34.5 <sup>b</sup>	35.2 <sup>b</sup>
	Fatness traits								52
15	Average back fat depth, mm	7.1*	8.7**	0	10.4	27.2*	27.7³	25.5 <sup>b</sup>	24.7 <sup>b</sup>
	Weight of internal organs Heart, gram	***/6	11.4***	0	14.4	2268	225*	238 <sup>b</sup>	244 <sup>b</sup>
20	<u>Meat quality traits</u> Reflectance value, EEL	5.7	6.1*	_	<b>8.1</b>	18.64	18.4ª	21.8 <sup>b</sup>	19.7
	*P<0.05; **P<0.01; ***P<0.001								PCT/EP99/10209

## Table 1, continued

¹Only the traits for which the QTL peak was in the IGF2

region (0-10 cM) and the test statistic reached the nominal significance threshold of F=3.9 are included.
²"QTL" is the test statistic for the presence of a QTL under a genetic model with additive, dominance, and imprinting effects (3 d.f.) while "Imprinting" is the test statistic for the presence of an imprinting effect (1 d.f.), both obtained at the position of the QTL peak. Genome-wise significance thresholds, estimated by permutation, were used for the QTL test while nominal significance thresholds were used for the Imprinting test.

<sup>3</sup>In cM from the distal end of 2p; IGF2 is located at 0.3 cM. <sup>4</sup>The reduction in the residual variance of the  $F_2$  population effected by inclusion of an imprinted QTL at the given position.

5Means and standard errors estimated at the IGF2 locus by
20 classifying the genotypes according to the population and
parent of origin of each allele. W and L represent alleles
derived from the Wild Boar and Large White founders,
respectively; superscript P and M represent a paternal and
maternal origin, respectively. Figures with different letters
25 (superscript a or b) are significantly different at least at
the 5% level, most of them are different at the 1% or 0.1%
level.

Table 2 Maximum likelihood phenotypic means for the different F2 genotypes estimated under (i) a model of a mendelian QTL, and (ii) a model assuming an imprinted QTL.

	Mendelian QTL				Imprinted QTL		
Traits							
	µ <sub>LW/LW</sub>	μ <sub>LW/P</sub>	μ <sub>Р/Р</sub>	R	µ <sub>PAT/LW</sub>	μ <sub>PAT/P</sub>	R
BFT (cm)	2.98	2.84	2.64	0.27	2.94	2.70	0.27
% ham	21.10	21.56	22.15	0.83	21.23	21.9	0.83
						5	
% loin	24.96	25.53	26.46	0.91	25.12	26.1	0.93
						4	
% lean	65.02	65.96	67.60	1.65	65.23	67.0	1.67
cuts						5	
Q O	6.56	6.02	5.33	0.85	6.43	5.56	0.85
backfat							
% fat	28.92	27.68	26.66	1.46	28.54	26.9	1.49
cuts						9	

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### CLAIMS

- 1. A method for selecting a domestic animal for having desired genotypic properties comprising testing said animal for the presence of a parentally imprinted quantitative trait locus (QTL).
- 5 2. A method according to claim 1 further comprising testing a nucleic acid sample from said animal for the presence of a parentally imprinted quantitative trait locus (QTL).
  - 3. A method according to claim 1 or 2 wherein in the pig said OTL is located at chromosome 2.
- 10 4. A method according to claim 2 or 3 wherein said QTL is mapping at around position 2p1.7.
  - 5. A method according to claim 1 to 4 wherein said QTL is related to the potential muscle mass and/or fat deposition of said animal.
- 15 6. A method according to claim 5 wherein said QTL comprises at least a part of an insulin-like growth factor-2 (IGF2) gene.
  - 7. A method according to anyone of claims 1 to 6 wherein in the pig said QTL comprises a marker characterised as nt241(G-
- 20 A) or as Swc9, as identified in figure 4.
  - 8. A method according to anyone of claims 1-7 wherein a paternal allele of said QTL is predominantly expressed in said animal.
  - 9. A method according to anyone of claims 1-7 wherein a
- 25 maternal allele of said QTL is predominantly expressed in said animal.
  - 10. An isolated and/or recombinant nucleic acid comprising a parentally imprinted quantitative trait locus (QTL) or functional fragment derived thereof.
- 30 11. An isolated and/or recombinant nucleic acid comprising a synthetic parentally imprinted quantitative trait locus (QTL)

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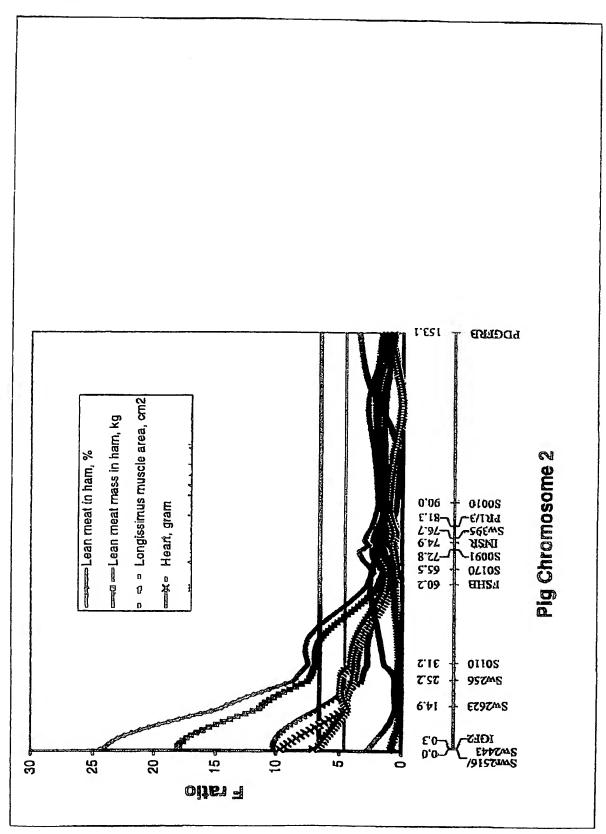
WO 00/36143 PCT/EP99/10209

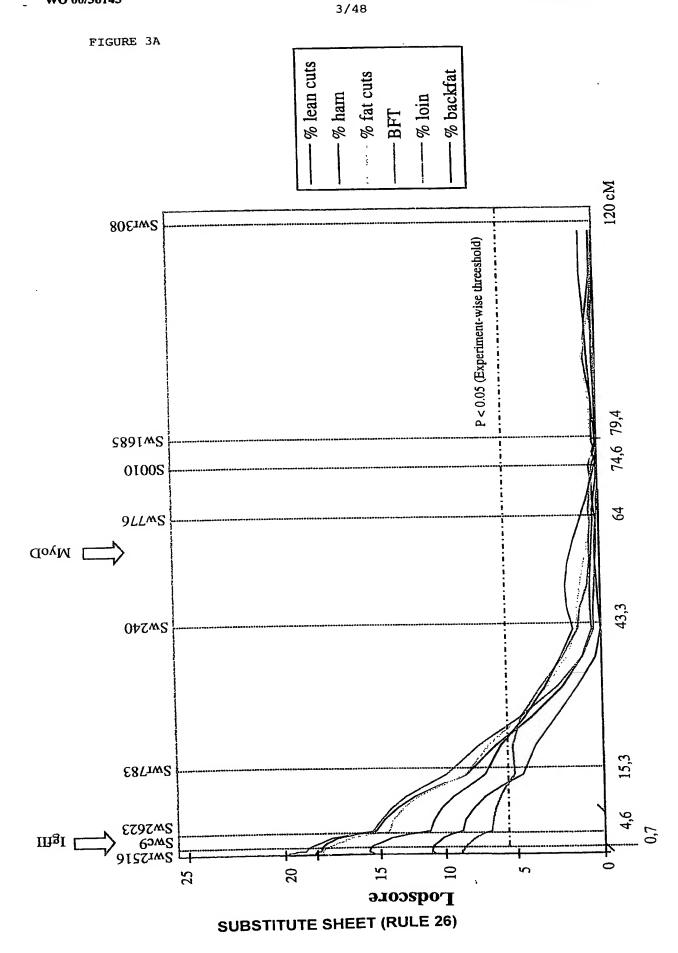
derived from at least one chromosome or functional fragment derived thereof.

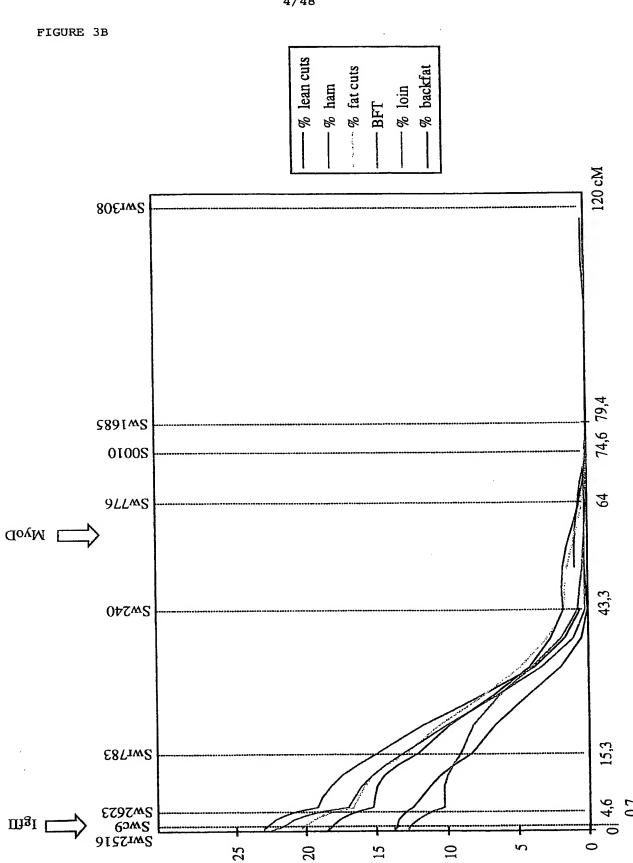
- 12. A nucleic acid according to claim 10 or 11 at least partly derived from a Sus scrofa chromosome.
- 5 13. A nucleic acid according to claim 12 wherein said nucleic acid is at least partly derived from a Sus scrofa chromosome 2, preferably from a region mapping at around position 2p1.7.
  - 14. A nucleic acid according to any one of claims 10 to 13 wherein said QTL is related to the potential muscle mass
- 10 and/or fat deposition of said animal.

- 15. A nucleic acid according to any one of claims 10 to 14 wherein said QTL comprises at least a part of a insulin-like growth factor-2 (IGF2) gene.
- 16. A nucleic acid according to anyone of claims 10 to 15 wherein a paternal allele of said QTL is capable of being predominantly expressed.
  - 17. A nucleic acid according to anyone of claims 10 to 16 wherein a maternal allele of said QTL is capable of being predominantly expressed.
- 20 18. Use of a nucleic acid or fragment derived thereof according to claim 10 in a method according to anyone of claims 1-9.
  - 19. Use according to claim 18 to select a breeding animal or animal destined for slaughter for having desired genotypic or potential phenotypic properties.
  - 20. Use according to claim 19 wherein said properties are related to muscle mass and/or fat deposition.
  - 21. An animal such as pig selected by a use according to claim 18 to 20.
- 30 22. A animal according to claim 21 characterised in being homozygous for an allele at a paternally imprinted QTL, preferably located at a Sus scrofa chromosome 2 mapping at around position 2p1.7.
- 23. An animal according to claim 21 or 22 wherein said QTL is 35 related to the potential muscle mass and/or fat deposition of

FIGURE 1







Lodscore SUBSTITUTE SHEET (RULE 26)

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FIGURE 4

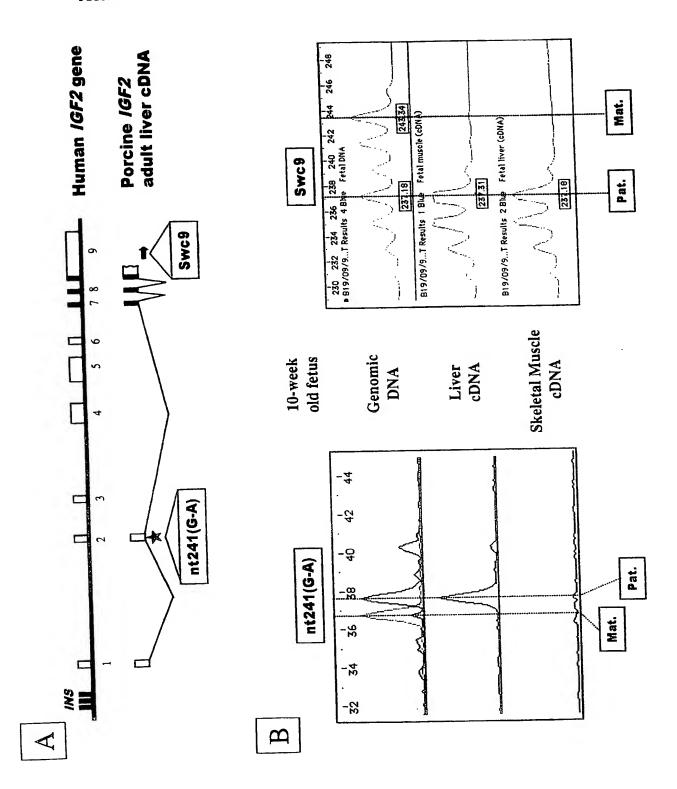


FIGURE 5

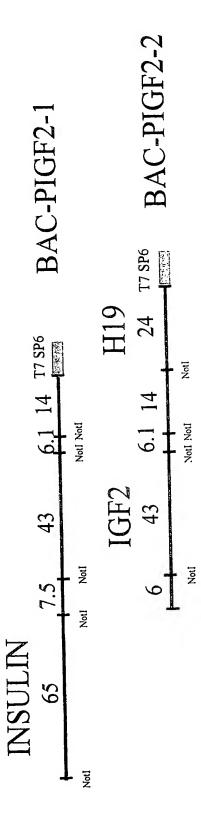


FIGURE 6

Contig 1 (500 bp)

Contig 2 (943 bp)

Contig 3 (1500 bp)

GGGGAGGGATGCTCAGACCCGCTCTGGGAAGAAGAGACCCTCAGAAGAATCCCTTCCCAAGGGTCACGCGG TGGAGCCCAGGGGCCCGCTAGGGGCCGATTCCCACAGCTCGTGCTGCCACCTGCTGGCGCTCCCAGGAACTGC GGAGGCGGTGGGGGCCCTGGATGGGTCCGGCAGTGGGCTCGCAGGAGACCCCTGGAGGGGCTGCGGACACCCC AGCTGCCACTCACAAGGTGCCCAAGCGGCGGTGGCAATGGGCTGAGCCTCTCCCCCCCTCCTCCTCCGCAGGA CATTGGCCTCGCATCCCTGGGGGTCTCGGACGAGGAAATTGAGAAGCTGTCCACGGTGGGTTTCTCCCCCTGC

AGGGCCCTGGGTTCCAGCCAGGCCCTCCTGTCCAA

GGGGGACCCCTGCTGGGGGATGTGGGTGCACAGCCAGGGCCACCAGGGAGTCAGGACACGGGGCTCCCTTCCC
TCGGGTCCCTGAGACCCCTGGCCTCCCGCCAGCACTCCCTGTCCGAGGAGCCCGAGATCCGGGCCTTCGACCC
CGACGCGCGCGCCGTCCAGCCCTACCAGGACCAGACCTACCAGCCCGTCTACTTCGTGTCTGAGAGTTTCAGT
GACGCCAAGGACAAGCTCAGGTGGGCCGGGGCCCCGGGGCCCCCAAACTGGAGGATCCAGCCCTGCAGCCCGCC
TATGAGCCCATTTCCCAGCAGAGGGAGCTGCTGCGGACCCCACCGTCACAACCCCCCTCCCACAGCTGGAACC
CCAGAAAGCCTGCGGAGGGGGGACCTGCAGGGCTG

Contig 4 (3024 bp)

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FIGURE 6, CONTD.

AGGCGGCTCCAGGGAGGAATCTTACGGAGTCAAGGCCCGGGTGCCGCTGGTCTCCGAGTGACATGGCCGTGGT GTCCCRTCTGCCGGCCCACATGCCCGTGAGAGAWGCCCCATCCCCCTGGGAGGGGGCCCCGTGCCGGGCAGGC GGCGGGAGGCCCAGGACCGGTGGCTGCTGCGGCTTCCACTCCAGGGTGGGCGGGTGGGGGGGTGGCTGTCTCT GTGTGACCGGCTCTCCCCGCAGCAGCTGCCGTGGAGCTGGGCGGAGGCCTGCAGGCCCTGCGCGC  ${\tt TGGAGGGGCCCCGCAGAAGCGTGGCATCGTGGAGCAGTGCTGCACCAGCATCTGTTCCCTCTACCAGCTGGA}$ GAACTACTGCAACTAGGCCGCCCTGAGGGCGCCTGCTGCTCCCCGCACCCCAAAACCCAATAAAGTCCTGAA GGGGGGCCTGCCTGCGACCCCTCTCTGCTCTCGCCACATCGGCTGCTCTAAGCTTCCTCCACATGCATCGGGT GCCCACAGGCACATGGGCACCGGGGGACCAGGGCCCAGGGCAGGGCCCTTCAATGTGGCGAGCTCTGGTTTTC CCCCGCGGGACCAAGACCTGGCCAGCCTGCCAGTCGCCCAGGCCAAACCAATCTGCACCTTTGCTGAAGGTTC CACCGGGCCAGCACTGGGGGGGGCGGGCCTAGAGCTGGGCGCCCGGGGCCCCAGGGACTGCACCCGCCAG AGGCTCTCACCTGTGTCGTCCCCTCCCCACGGCCACACAGACACCCCTGGGGAAAGTCACAGGCCCCCAGCA CTGAGCTTGTGGTCCTGCCAACCAGGGAGACCCGTGACCACCCTGCTGCTTCCCCTCCCCCCAGGGCCAGCA GACTCCTTTGGGACTCGGGGCCCCTGAGCCGCCCCCACTCGCAGGACTCACGGGGTGTGCGGTCCTGGGTGAG TGGGGGCTTGGGAGAGGGTCACTCTTGTCCGTCGGGTGGGGAAGGCTGAGAGTCATGGTGTGACAGCGCCCTC GGCCTGCCGGGTGGGGGGTCTCCCTTCTCCCGAGCCCAGATCCCCGGGTAC

#### Contig 5 (1730 bp)

CGTCACCCGCAGAAGCCAGGCCCACAGGCCTTGGCTCAGCCCCTCCACCCAGGCCCACGTTCCGCCCCTTCTG
GGAACTGGAGGACAGCCCGCCCTCGCCCTCGGACCTGGCTTTGTTTTGCCCTGGCATCTGGCAGTGGCCGGCAG
CTGCGTTCAGCCCTGGATGACACCCTGGCGTGAGCGGTGGGTCCCCGTGCTGAGGGCAGCCCCACACACGTC
CTGCTCACTTGCCTTGTGTCTGCTCCGCATCCCGTCATCACACATGCCATGCTGGGGCACCGTAGCGCCTTGC
CCTGTGTGGCACTGTGGCACTGTTTCCTGATGGGAAGACTGAGGCTGGGGTCAGGCCCGCTGCCCACCC
TCTAAGGACATTCTGCCGGTGCAGCTCCCTCCAGG

ACCAGGCAAGGTGGTCCGAGCGGTCATTCACAGACAGAACCAGCAGAGGGCGCCAAAGCCCCACTTTTGACAA
ACTCCCTTCGCCCTGAGCCGAAAGTCCAGGCGGCAGGTGGACCTCTCTGCAGGGCTCTGCCACCCCTGCTGC
CGCTTGCCAGCACTCACAGGGGCTGCGGGGGGTGCCCAACAGGCCGGCTACCCTGAGCTCTGGAGGCGATGGA
GTTTAGGAGGGAACGAGGGGACTCCTGGGGGTGACTTTCTTCAGCGCCCACATTGCGGCCCAGCAAACCGAGG
CTGGAGGAGGCCGGCACCTGTGCCCAGCTGGAGCCTTTGCTGAGGGTCTCCAAGGCCTGGGGAAATTGAGGC
TGGGGGCTGGGGGGTGTCACTGTCGGGCCAGGAGG

ACTCGGCTCCAACCTCCCGCAGGCCCCTGGCACGGTCTCCAGGAGTCCACTGAGGGGTCCCCAAAGCTGCCACCAGGAGCTGGGCCTGGGTCTGTCACCACCCCCACCCTCCAAGTCTGAGATATG

Contig 6 (4833 bp)

ATGTGAGCTGCACAGCATGAGCCCTCGGCCCCACTGCTGTGGCCTTGCGGACATTGAGGTGTGTGCCGCCCAG GGCGACCACACCCTGGCCTCTCAGGGTGCCCGTACAGAGGCGGCTGGGTCGTANGAGGTGCGGGGGCTCTGGGG ACCGCTGGTGAGTTCAGGACGGGGGTCATGCCACCTCCTCTCTGAAGGTTTGGTGAGGTGGCCCTTCTCTTAT CGTGATGACAATACTGATTTCTGGAAGAGCCAGGTGTTTTCTGAGGCTGTGCTTGCACTTCTCCACGTGGCCA CAAGGTGCCGGGCTCGGGTCAGATTTGAGAAGCCCTGCGGGAGCGGGTGTCATGCGCCAGATTCAGCTTGCCT

CCTGCGGGTCTGGGGTCAGGACGTGGTCCCCAGCAGTCTGCTCCAGAGCCTGTCAGTGATGTGTGGGATTTTA TGCTGCANNGCCCCCTGCCACCTGGTCGGAGCCNCAAGACGGCATCTAAAGATCAGTTCCTCATCATCAGTTC CGCAGTGCTGGGGTGGGGCAGATGAGAACCTCAGGGCTGGGCGCAGAGGTGGGGAGCCCGCCTGGACCCCGA CACTGCAGGGGGGCCTCCCCCTTGTAGGAAGAACAATGTCGCTTTGCCACCCAGCCCTCTCCCCAGGGTGCCC CGAACTGTTGCTCCTAAGACCTCTGGGCTGTGTGCTGTAATTCTATAAGTGGCCACCAGGTGTCAGCAGGAGG CCACTTAAGCATCCATGTGGCGGAAACCTGGAGCTGGGGGTTCCTAAGGGTCCCTCGAGTGTCTCCTGAATAA ATAGGCGCTGACCTGATCCCCAGGAAGGGATAACCCTCTCCCAGGCCTAAGAGGCAGTGGGGCAATGAGGTTT ATGTGTCCACTGTACCCCCAAATTGTCTCTTCCTTCCCTCTACCCTGTGTCCCCACCGTGGACGATACACGGA GTGCGAGGCTGCGGGTCACAGCCCTCACAGCCCCAAAGCTGCAGGTCCTGCCTCAGGGGCCACCGCAGCTTGGC TGGTCCCCCTTGGGTCCTCCCCACCCTGACCCGTCCTCTGCTCCCCTTTGCTTAAATGCTCTGCGTTTC AAGGTTCTGATGGAATAAAATAGCCCTGCACTGGTGTGTTCCTCTTTGGGGCTGTGCCAGAAGTGGGAATTCA GACCAGGGCAGAGCTCAGATTCCACATACTGTGTTAGGGATGGCAGGTGCCACATTTCCAGGAGTTTCATTGG TGGTTTGTAAATGCTACTTCCGTTTCAGCCCCTCAGCTGCCCACCTCCTCAATTTAGGGACCCCCCCTTTGG CGGGTTGCCCATGGAACCACATCATCTGGCGTGGGGTGAGCCCTTTATCCTCCCTGGCCCCACTGGGAGGGTT TGGGGAAGTCCCAGCTAAATTTCTCCGTAGGGACCTGGAAGGAGCCCTTGTGACATCTGGGCACAGATAAGAG GTAGGGGGCACAGGCCGTGAACACTTGAAGCTGCAGAGCCCAGAGCAGAGCAGCAGGAGCAAGTGACTGCTC CCCACCCAAGAACTGTGGGCTGCGTCACACTCCCCACTGTGTGCCCTGGACCTGACAGGGCCTTTAGCCT CCCTGCATCCCTCCCACCCAAGAACCCAGTGAGGCACCCCACTTGCCCCTCCTTAGTGTTGTTATGGCTCTG GGGCATCTGCATTTTGTTTAGGACACCCCCAGCTAGATTTAAGTCCCCCCAAGTGTGACTCTTTCCTCCACTG AAAACCCTGTCCTCCCACCAAAGGGCCCTATCCCTTTAGCTGAGCCAAGGAAATTCAGGAGGGCCCTTGAATG GGGGTGCAGTGAAGGTAGCGGCTGGTGGCCTTCTGGAAACTACATGTGACTTTGCCATTAGGTGAGTCTTTGC TTTGCCCCTGCTCTATCTGCAGGCTTATGGAAGAAGTTTAAATTCCCAGGGACACTTGGTCTAACCAGGCAGC GCTTGTATCTGGGCCCTTCCCCAGCTGCTGACCACTCTGAGTCTGCGCCTTAGTTGGAGTTTTGGCCAAGCTC AAGAGGCTGTGGACCCCAGTCATCCCACCCAGGGGTGCCTGTGGGCAGGACGCTGCTGCCTTTGCTGC AGTATTGTCACTGTCCGGCACCACACACATGGTGCAGGGGGTGGTATCAGGTGCCACTGGGGAAGGGAAAAA CTCCCAGGTGAGTCCCCTGCCTCTGGAAGCAAGATGGACATGACCGCACTGTGTTGCAGCTGCATTGGGAGGC CCCGAAGAAAGATTTTTCTGATCTTTCCGAACCCTGCTTTTCCCCATCATGCCCCGCCCCCATTTTACCCGT GCCACGCCCACTGGTGTGCCGGGGTGTCAAGTGACTGACAAGTGTCAATCTACTGAGGCCCTGCCCACTCTCC ACCCCCCACATAGTCCCACCTCCCAGCTGGCAGGGAGAACTTCCAGCTAATGCCCATGCCCACAAATGTCTT TCTGTCAGCCTAGAGCTGGACCAAATCTCCACCCTGTAACATGCTGTGCCCTGGCGTGGGAAGGTGCCAGAGC CAGTTGCCCCAGCAGCCCCAGAACCACTAAGTTGGCACAAAGCTACCCAAATTTGGAGGGGCTTGGGGAAGGG CATGGAGGGGATGAGGGGGGCAAAACTAATTTCAGTTAGCATTTGAGCAGGTGCCACGCTCAGCGTG GAGAGGCTCTCTTGCTTCTAGGGACCCATTATGATGCACACGCTAAAAGCGCCCTTCACCATCTCCCAGCCT CAGCTTTGTCCCCCTCCTCCTCAGCGGCAACCCGGCTGGAGGGTCTGGCCACTACAGCCAGAGCGCCCCC TACTTTGGTGGCGACTGCTACTATTGGCCCAACCAGCGGATCACCGGCCAGGCAGTTTCGGCAGAGAGTCTGG GGCACCAGTGACTCCCCCGTCCTCTTTATCCACCACCCAGGAGCTTCAGGGACTACACAGCGACTAGAGGGCA GGTAACTGGTCTGCCCTCGGGGCTGCCCCCTCAGAGTGTGTGAGAAAAGCTGCATTGAGTGTTTGGGTGC AGGTGGGCTGGGGGCTTGGGGCAGCCAACAGGAACGGCGGGACCTCTGCTTCCAGAGGACCCCAGATCCTGGC AAGCTTCGACTTTGGAGGGGACAGGAAAGACAGGTGGAGAGGGGACACTTCCCTCTTCTGTACAGACGCCCAC CCGGAGCCACAGAGGCTTTTGCAAGGAAAATAGGTTTTCCCTCACTAATGCAGCAGGCAAAATGGGAGGGCCA GGGGTGGAGGGTAGTGCCCCCCCCCCCCCAGCAGGAGGGCACAGCTGTTTCTGCAAATGTAAAAAAAGCAGGGTTT TTCTGTGTGAGAAGTTCCCTCTTGCTGCATGTCCCCACCCCGCCACCAAAGACAAACAGGACACTGTGCAGA GGGGCCAGAGCCCCGAGATTTTGGAGTTGTTTTTATATGCATATATACCATTTTGAAAGCAAAGCTTCCCTCT CCCCTACTCCCTACATGTCCCCCTTCACCAAAAAATCCCACCACGTAACTGGAAAGGGGGGGTGAGAAGGACGA CGAAGGGGCACTGTCCCCTCCCGTCCCACAGCGGGACTTAAAACGTACAGCTTTTCGCCTCCGGACAGTGTGC CGCCCCTGGCCCCGTCACGCTCCCCTGCCCGGGGGGCTGAGTGTGGGGCCAGGGCCTGTCTCCAGGCATGC ATTATTTTGTGCATGAAGGTTTTGTCCCGCCCACCCAGGCTGGTGTTGGGGGGGAAGGGGTTCATTGCTCCAAA GAAGCCCATCTCCCCCTCAGCCACCTTCAGCCGCCTTCGCAAGGCAGAGCTGTGTCCTCTGCTGTGTGCCTG CCCCGAGGCAGGCATTTGTGTGCGGCCCCCAGCCCCAGGCCCAGGCAGATGGGCCAGCCTGCCCGACAGA ACACTCTTTATTTTCCCCAGGGGCCGAAGAGTCACCCCTGAACTTGAGGACGAGCAGCCGGATTCCAGCCCCC AGCCCCAGGCCCCACATCTCCTCGGGCTCAGCCGCGCGCCCCAGCTGCCCCCAGCCTGAGCTGCAGCAGGC CAGGGCTGCCCGAGACCCCCAGGCCCCAGGTGAGCTGCTGCAGCCTGTGGCCCAGGAGATCTCCGCCGGCTCAG CGGCACAGAGGCTGTGCTGCAGGCCCAGACCTCCCAGGCCGTTTTAGTTCCCATCTCCCCTTGGGGGAGGGG TGGGGCTCAGAGGGGCTGCGTGCATCCGCAGAGCTGGGGTGCAGGGGCTCCAGGTGCCTCTCTCCCAGGCGGC TGGCCCGGAGGGGG

Contig 7 (2014 bp)

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FIGURE 6, CONTD.

CCCTCGGAGTTGGAACGTGGCTTCCTAAGCCTTCACCAAAATTGAGGCTTTCCGCGCATGGCGCGTGATGCC
CTTGCTGAATCAGAAGCACTCTGCCCTCTGATTCCTGCTTTCCACAACCCTGAGAGCATGATTTCTGGTCCCC
CAAACTCACTGAGCAAAAATCTTTTTTGTGGGGGCTGCAAAGATAGGAGGCATTTCTCTCCGGAGCTCTCCAAA
CTCCCTTGCCTATAATCAAGTTCCCTAAAACTTAGACAGAGCTTCCCAGGCCCCAGAGGCACACAGAGCCATT
ATTGGAGCTGCGTTTAATGATGACAGGGACCATGGGTCATGCAGCTCCCCCAAGTCACAAATGCCCCAGGTAT
CCTTGGCTCCAGCCCAAGCCCAAAGCAAACTCTTGC

ACACCGGTTCTTCAGGACCCACCGCCTAGACGGTCTTAATCCCTTTTTCCCCCAGACCTAGATTC

Contig 8 (371 bp)

Contig 9 (2415 bp)

CCAAAACTGGGGCCCTATCTTACTAGGGTTCCCTAAATGCAGACAGCGCCCGGGAAAATAGGGGCGTTTTTTT
TCCTGTTTTGCCAAAAATAAACTAATTGAAACCAATTTTTAGAATTAAAATCTAAAATGACCTTGATTTTCTGC
GTTCTCCAAATGTACTTTTCACAGCCCAGGTTGCCCCCAGTTTAGACGTGTTGCTTGAATCTCTAAAGCACC
CTGAGGATTTTTCCCGAGGAAGCCACCACAACTACGGAATTTACTGTCCTTCGGGGCCACAAGCCTCCAGGCC
ACCAACTTGGATTTCTAAACCGTGGAAATCAGCCTCCACTTCCCTCCGCCACCCCGAGGGTCTGCTCAGACCC
CCCAAACGTGCCGCTGTTCTTCTCCCCCCAAATT

ACAGCATACGTTTTT

Contig 10 (3753 bp)

AGATTCCAATGGGGATCCCGATGAGGAAGCCGCTGCTCGTGCTCGTCTTCTTGGCCTTGGCCTCGTGCTG CTATGCTGCTTACCGCCCCAGTGAGACTCTGTGCGGCGGGGGGGCTGGTGGACACCCTCCAGTTTGTCTGCGGG GAGGACCTCTCCCGAGGGTCTGAGACTTCAGAGCGGGGGGCGCCCTGGCCCTGCGCAGTGATTGGCACCTGC CATGTGCCTGGCTGGGGCTCACACCCCCTGACGTTCCTGCAGCGTGACTCGAAACGGGAAACCGAAGGGACGG AGGCCCCACAGGATGACAGCCTGTCCCTCCTGCTCCTTGACCTGCCCACAGCCAGGGCTGCAGGCACTG ACATTCACCCATGGTATTGTGGTGCCTGACGTCTTGGCAGTGGGCATGGGTTCATGGACTGTTGGATTGAAAG CACCTCCCAGCAGGCTGGGCCTCAGTGTCCTTACCTGTAGGATGGGTCAGGGGGCGTCCTGGAGAGAGTCCTCG GGACAATGGGGAGGCTGGGGGCAGGCCTGACCCTGAAGGTGGGAGTGTGTGCTCCCCCTGGGCTCAGC CCCTGGCCTCTGAGCCCTCTCTCTCCTGCCTCGTTTGGGGGCAGGGAGTGGCACCATAGAATCTGGCGCTGGG CCTGGGGAGCGGCCCCCTCGTGCCAGGCTTCCCCGAAAGGAGGGCTGGGCTGAGCTCCCGACCCTCTGGACCC CCTCACTCCTCCCGGTCTTCCTCCTCCTCCACTTCCCACCTGTGTCTCCGGGTCCCGGGGCCGCAG GCTGCCCAGGCGCCTGCTGATCCATTGGGGACCGCACTCGGGTCCCCGCTGGCCTTCGGGTCAGGGCCACGGC CCTCCCCTTGGTCCTGTGGGACTTCCAGGCAGGCCGGCAAGCCGCTGAACCGCCGCAGCCGTGGCATCGTGG AAGAGTGCTGCTAGCTGCGACCTGGCCCTGCTGGAGACCTACTGCGCCACCCCCGCCAAGTCCGAGAG **GGACGTGTCGACCCTCCGACCGTGCTTCCG**GTAAGGCAGCCCCTCTCTCGGCAGCGCCCCCCCGGGGGGG GGCTGTCTCCTCTGAGCCGGGGGACCGGGCGCGCGCCCTCTTGGGCCTCCAAGTGCTGCCAGAGGGGCCTTC ACTTCCCCAGATACCCCGTGGGCAAGTTCTTCCGCTATGACACCTGGAAGCAGTCCGCCCAACGCCTGCGCAG GGGCCTGCCGGCCTCCTGCGCGCCCGGGGGTCGCACGCTCGCCAAGGAGCTGGAGGCGGTCAGAGAGGCC AAGCGTCACCGACCCCTGACCGCCGTCCCACCCGAGACCCCGCCGCCCACGGGGGGCGCCTCTCCCGAGGCGT CCGGCCATCGGAAGTGAGCCAAATTGTCGTAATTCTGCGGTGCCACCATCCACCTCGTGACCTCCTCGACC CCGTGCCCCAACCTCCCCATGTCAGGCTAGTCTCTCCTCGGCCCCTTCCATCGGGCCGAGGGCATCCAAACCA CAAACCCAATTGGCTTGGTCTGTATCTCCCCCCAAATTATGCCCCCAATTATCCCCAAGTTACATACCAAAAA TTGAACCCCTCAACCACACCCACATACAATCAGCCCCCGTAAAACGAATTGGCATCTTTAAAACACCAGAAAA AATTGGCTGTGACCCATCATCCAAGAGAAAGGAAGGGACCAAAATTTGCAGGTAGGCTTGTCGCCGCTCACAG CCATCTCCCTCCTGCCACACCCTCGCCGGCCACTGGCGGTGTGGCACCAAGGACCCAGTCCCGTCTCTC TCTAGTCCCATGACCGAGACCGCGGTGGAGTTGGCTGGGAGACCCCGTGAGATCAGAGGAGGGGGGGAGCACGGAA CCAGAAACCCAAACCTGCACAGGTACAACATGACTGGCCCCCGCACAGCCCCAAGACCTCTCATCTCAGTCTC 

Contig 19 (500 bp)

#### Contig 22 (450 bp)

TGCCAGAGACCTCAGAGCTGGGCTCTCCCTGCCTGACACGGAGGG CTGTGGCTTCCACCACCCCAGGCCACACCCAGCCTGCCCAAGTCCCTGAA GTGTCCCCAGAGGTGGCCTGCCTCCACGCCCAACATCAGGCCTGCTGCA GCCCTGGACGGCCCCCTGTCCCCCGGAAGCCCTCGGGGCTCTCTCGCGTC GCCTCTGGGGAACCCTCGGTAATGTGGCCCAGCCGTGCAGTGGCCGGATC ATTTGCTCAGGGGGGCCCAAGGCAGGGGGGTGACACATCCGCAAGTACCG CATATGCACAGGATATGGATTGGGTGTGGATTTAACCTTTTCGCAAATGT CTCTGCCGGTACAAATATTGTTTCTAATCCTCTGCCTCCCTGAGCCGGTG AGTCTGCCCGGGAGCTGCGGGGAGCTGCTTGCTGAACCTGCCCTGGCCC CCACCCCCAAGGGAGCCCCCGGCCAGTGCTGAGGGCAGGAAGCTTGGGCA CAGGCTGCAGAGGCCAGCGCTGGCCTCAGTCACCT

#### Contig 24 (868 bp)

CAGGACGCANGTGGGCGTGTGTGAGTCCGTCTACACGTCCAGCCAAGGGC
GGCCGCGACCGGCCAGGGTGGGCAGCCCCAGCCTCAGCAGGGCGCTCTCT
GGGGCTCAGGCTGCGCCGACGGGAGATGAGGGGTGAGGCGCAGTCTGGGG
CTGCTGCCGCAGAACCTCGCCCAGCTGGCAGCTGGGCACAGGGAGACCTG
TACTCCCAGAACCTGAGGCTGGACGTCCGAGACCCGCGTGCCGGCCTCTT
GGGTGCCTGGTCAGGGTCCTCTTTCTGGTTTGTGGGCAGAACCTCCTCAG
CGCGTCCTTGCATGGGGTGCTAATCACGGAGTAAGGAGCCAGAGAATGAG
GCACGGAGTATCCAGTGTTAACCCTGGAGTATGGAGACGGGAGTACTAAT
TGTGGAGCATGGCTCTAAGGAATGGAGTATTCGTCACGGAGAACGCGGGG
CCGGGTGAAATACGGAGAGCGGCGTACGGACAACGGGGACACTAGAGA
TGTATANNGGGCGTCAAT

#### Contig 25 (500 bp)

ACCAGTTTCGATGAGCAATCCCAGCGGCGTAACATTATGGCTGCAGCCTG
GTCAATGCCGGTGGAGTTTGAACCTCCACGCGTGGCGATTGTGGTAGATA
AATCGACATGGACCAGGGAGTTGATTGAACATAACGGTAAATTTGGCATC
GTTATCCCGGGCGTTGCAGCAACTAACTGGACGTGGGCGGTGGGAAGTGT
GTCGGGGCGTGATGAAGATAAATTTAATTGCTATGGCATTCCGGTTGTGA
GAGGCCCGGTATTTGGTTTGCCTCTGGTCGAGGAAAAATGTCTGGCGTGG
ATGGAGTGTCGATTGCTACCTGCGACTTCTGCGCAAGAAGAATACGACAC
GCTGTTTGGCGAAGTAGTATCAGCAGCGGCAGACGCACGGGTATTTGTCG
AAGGCCGCTGGCAGTTTGATGATGATAAGCTCAATACGTTGCATCATTTA
GGTGCTGGGACGTTTGTTACCAGCGGCAAGCGTGTTACGGCGGGTTAAGC
Contig 26 (900 bp)

ATGTTTGATGTCCGCGCGTGCTGTAAAAATTTACGCTGCTCGCGTTCTTT GGCTTCGTCCACCACCGGAAAACGGACAAAATTTCCGTCATACCTTTTT CTTTCAGGCGGAAGCCAATGTCGTAATCTTCAGTAAGACTCTGCACGTCG AAAGCAATACCGTCACCGTCAGCTAACAGTGCGGTCACGGCGCGGCGGCT GAAACAGGTGCCGACGCCTGCGCTGGGCACTTGTCCGGCGAGGGCTTCAC GCACCGGAACATCTTTGCCATGCAGCTCTGAAAACTCATCAATGTAAGTC ATGCTGGTGAAGTGCGTCCATTCGCGTTCGAACGGATACACCGGGATCTG AATCAGATCTTTACGCTCGACCAGATAGTTGAACAGACGCAATTCCATCG GTGAAATCACATCTTCGGCGTCATGCAGAATAAAACCAGCAAAAGCGAAA TTGGCGCTACGCTCAAATTGGGTGATGGCGTCCAGCACGTTGTTCAGACA GTCGGCTTTGCTGGTGGGGCCAGGCCGCGCGCAGACTACCTTATGCACAT TCGGGAAGCGACCCACACTTCGTCAACATCACGCTGAGTATCGGGGTCG TTGGGGTAGGTGCCAACAAGATATGATAGTTTTCGTAGTCGAGCGTGGT CGCCGCCAGCTCGGCCATATTGCCGATGACGCCCGTTTCATTCCACGCCG GAACCATAATCGCTAACGGTTTTTCATCTGGTTTATACAGTTCGCGGTAA CTCATTCGCGGGTAGCGGCGATAAACACTCAACTTGCGTTTAATGCGGCG TACCCAGTATACGACATCTATAAAAAAATCGTCCAGCCCGCTGATGAACA TGATGACCGCTAACGTTATCGCGATTACTTTTAAGCCGTATAGCCAGGTA Contig 27 (500 bp)

AGTGCGGTCGGCCGTCCTGACGCTCAACACCGTATTTCCACGCGACCGC GGATTCAACCTGGTCACACGGACGCCATGTAGACATGTTCGGGGTTACGC GCAGAGAAGCGACCTGCTCAACCGGCTGGTGAGTCGGGCCGTCTTCGCCC AGACCGATGGAGTCGTGGGTGTAAACCATCACCTGACGCTGTTTCATCAG CGCAGCCATACGTACGGCGTTACGTGCGTATTCCACGAACATCAGGAAGG TGGAGGTGTACGGCAGGAAGCCACCGTGCAGGGAGATACCGTTAGCAATC GCGGTCATACCGAACTCGCGAACACCGTAGTGGATGTAGTTACCCGCAGC ATCTTCGTTGATTGCTTTAGAACCAGACCACAGGGTCAGGTTAGACGCG CCGGGTCAGCAGAACCGCCGAGGAATTCCGGCAACAGCCGGACGAACGCT Contig 29 (450 bp)

TCAGGCCAATCTGTCTGGTCTCCAATGGGGACAATTTGGTTCTTTAGGCT
TCTGTCCAATGGTCCGAATGGCCCACTCCCCGGGCGCCCAAGGGTCC
TCTGTGCCTCGGGTGGGCTGGCACGGACCGCCCCCAGGGTCGTGCCAGCC
CCGTCACCGGGGCCCAGAAGCTTCGGGCCTCTAGCTGGCTAGTCGGGCTG
CTGTGCAGGGGGCTGCGCTGGGGGCAGAGGCGGGGGTGAGGTAAACCTC
CCAGCCGCCCGGGGTCCCTGCCGCAGCCCTAGGCGCCGAGACGGTGGCTG
GGTCGGTACCGCCAGACCCGAGGCCTCGGGGCCCGGGTGACCCCAGCTG
TCGCACACGCTCGCAGCTCTCTTGCTCATCAGGGCTCATCCCTCTGGACC
TCTCCTACTGCCCCACCTCACCCCGCCTGGACCCCATGAAGCCCCGCGGA
Contiq 30 (600 bp)

GCGCGGCGTTCCGGCTGGGGTATTTAACGTGGTCACCGGTTCGGCGGGC GCGGTCGTAACGAACTGACCAGTAACCCGCTGGTGCGCAAACTGTCGTT TACCGGTTCGACCGAAATTGGCCGCCAGTTAATGGAACAGTGCGCGAAAG ACATCAAGAAAGTGTCGCTGGAGCTGGGCGGTAACGCGCCTTTATCGTC TTTGACGATGCCGACCTCGACAAAGCCGTGGAAGGCGCGCTGGCCTCGAA ATTCCGCAACGCCGGGCAAACCTGCGTCTGCGCCAACCGCCTGTATGTGC AGGACGGCGTGTATGACCGTTTTGCCGAAAAATTGCAGCAGGCAATGAGC AAACTGCACATCGGCGACGGGCTGGATAACGGCGTCACCATCGGGCCGCT GATCGATGAAAAATCGGTATCAAAAAGTGGAAGAGCATATTGCCGATGCGC Contig 32 (450 bp)

ACGTGAGGTTTGGGGGAGGAAAGCGGGGGACGAGCAGCCCGAGAGGAGTG
GGGGCTGGCCTGTGGCTGATGAAACTCTGAGAAGGTTAAGAGCCCCCATT
TTTGTCTTCCTCTTTTTTATTATGGAAAATTCCAAATGGATGCAAAAGTC
CCAAACCTAACTGGACATCTTCTTGGTACCAGGAACGGTCAGGCACTTAT
GATGCACCGAGCCCCGAGGGAAAAACCCTGCCGTCCTGGAGCCCACGGTC
CAGCAGGGCACACAGGCCCCAGCCCGCAAGCGGCACGGCTGAGTCAGTGA
ATGGCGTGCCCTCTGGTCAAGGACGGCACTCTGGACCCCAGGGAAGCCT
CTGAGGAGCCCCCTTCACAGCGTCAAAAACTGTTAACAGGGCCATGTTCG
CACCCCCCCACACACGTGGTTCAGAAGCAGACCCCAGGCATCGTAATATG
TCATCCGTGAGTTCCCTGTGTGCCACCAACAGAAAGCCCATCGTCACGTT
Contig 34 (400 bp)

 ${\tt TGGCGGTGAACTATGTCGTGCGTGAAGAGCATTTGTGGTCGGTAGCGCGT}$ 

TATATGCGGGAAGTTTAGGCGAACTGGACAGCCTGGGTTTATCCGGTAGC
GAAATCCGCTTTCACGGTAAAACGCTGCTAGCGCTGGTGGAAAAAGCGCA
GACATTGCCGGAAGATGCCTTACCGCAGCCGATGCTTAACCTGATGGACA
TGCCGGGTTATCGTAAAGCGTTTAAAGCGATTAAGTCGCTGATTACTGAC
GTGAGCGAAACGCATAAGATCAGCGCCGAATTGCTGGCATCGCGTCGGCA
AATCAACCAACTGCTGAACTGGCACTGGAAACTGAAACCGCAGAACAATT
TGCCGGAGCTGATTTCCGAGCTGGCGTGGTGAGCTGATGCGGAAGCATT
ACACAATTTATTGCAGGAATATCCGCAGTAAAATCTTCCGAAGCCGGACT
GGGCGCGCTCAGCGCCACATCCGGCTTCGGCAAACTACAAATCCAACACC
Contig 36 (500 bp)

ATCCTTTTGGGGTCTGGCAATTACGCAATAAAGAAGGCCCCCATGCGATT
AAAGTCACCGGCCCACTGTCGTCTAATCATGGAGAAATTGTCCATCAGTG
GGGTCTCGATGGGCAGGGGATTGCTCTGCGTTCCTGGTGGGATGTTAGCG
AAAACATTGCCAGTGGTCATTTAGTGCAAGTGCTACCGGAATATTACCAG
CCAGCGAACGTCTGGTCCGTTTATGTTTCAAGGCTGGCGACGTCAGCGAA
AGTGCGGATAACGGTAGAGTTTTTACGCCAGTATTTTTGCCGAGCACTACC
GGAATGTTTCACTGTTGCATGCCTGATTTATGATTCAATTATCGGGTTGA
TATCAGTTTAAAACCTGATTTTCTCCTTTCTAAGCCGCTACAGATTTGGT
AGCATATTCACCTTTAATCGCGCATGATCTAAAGATAATTGAAGAGGTTA
Contig 39 (450 bp)

AATGTACTGGCAAAAAGCCAATGGCGAAGCGTGGGGAACGTTACATGCTC
TGCTGGCGGATATTAATAGTCAGGGTCAGGTGCAGATGGCGATGAACGGC
GGCATCTATGATGAAAGCTATGCGCCGCTCGGTTTGTACATCGAAAACGG
TCAGCAGAAGGTGGCGTTAAATCTCGCTTCAGGTGAAGGGAATTTCTTTA
TCCGTCCTGGCGGCGTGTTTTATGTCGCGGGAGATAAAGTCGGCATCGTT
CGTCTGGATGCCTTCAAAACCAGTAAAGAGATTCAGTTTGCGGTGCAGTC
AGGGCCAATGTTGATGGAAAACCGTGTAATTAATCCGCGTATTCATCCCA
ACGTCGCCTCAAGCAAAATTCGTAACGGTGGTTGGGATTAATAAACATGG
GAACGCCGTGTTTTTTGTTGAGCCAGCAGCAACAAATTTTTATGATTTTG
Contig 40 (400 bp)

GACATTAATCATTTCAAAATCAAAGCCCCGGTTTTCCATCGCCCGTTTGG
TGGCGTGGCACTGAACGCAATCGTTACGAGTGTAAATAGTAATGCGCATG
ATTCGTATTTCCGTTTAAAATGAAGATACGGCGCGGATGATACGCGTCGGG
TTGTCTCTCTGTTGATACAGAGATACTAGATGTAGTTGAAAAAAAGATTCA
ACCACACAATATATAGCCCAGTAGGGGTCGAAATTACCCTGGATATGAGC
GTGACGGGGTAGGGGGATTTTTGTGATTCACCAGGCAAAAAGAAACCCCG
AAGACAGGCTTCGGGGTCAAAGACGCGTATTTATTATCATTTTTTGCACTA
CGATTTGCGCATGCTTAACAGTGCGCCGATTAAAAATATCTACCGCAGCTG
Contig 41 (500 bp)

GCAAAATCACGTCCGCGACCTGGCGTTGTCGCTGGGCCATATTGGCAAAG GAGCTGGATTGCGGTGCCTGCAAAGTGCCCTGAATAATGCCATTGTCCTG TACCGGGAAGAAACCTTTCGGAATGAACACCCACAGCAGCACGCTAAGCA GCAGCGTGCTGAGTGCCACGCTTAAGGTCAGCCACGGATGATTCAGCACT TTCGCCAGTCCACGACCATAGGCGGCGATTATCCTGTCGAACATTTTTTC CGAGGCACGGGAGAAGCGGTTCTGTTTACGCAACGACTCCTGGCTGAGCA TCCGCGCGCACATCATCGGTGTCAGGGTCAGCGACCACCGCTGAGATC

AAAATCGCTACCGCCAGGTAATAGCAAATTCGCGGAACAGTCGCCCGAC GATATCGCCCATAAACAGCAGTGGGATCAACACCGCAATCAGTGAGAAGG TCAGCGAGATAATGGTAAAGCCGATTTCACCTGCGCCCTTGAGCGCCCCC Contig 42 (400 bp)

CCTGGAGGGCTCGGGAGGAGGATGCGCTCAAGCTGGCTCCCCGTGGGGC TGGCCGGAGTAGCCTCCGTGAGGGCACCGTGTCTGCTCCCAGAGCCCGC TCCCCGGCCTGCCTCCCTTCCCTGCCCCAGTTCCCCCGGAGCCCC AGAGCTCTGAGGCCACCAGACCTGGCCAGGACCCTTCGTGGGAAGAAGAG CTTCCAGGCGGGCTTCCAGGCAGGCCAGTGTCCTGGGGGCTGGAGGGA GTCCCTGGCTGCTGGGGGGCGCCGCAGGAGCACCTGGGGCGTCTGGGAAGAG AGCGGGAGAGACTGGAGCCAACTGGGGGGACAGAGGAGGGGTCCAACCC CAGCGGTGGTGGGGGGGTGCTGGTGGTGGAGGCCCTGAGAGGCTGTGCT GGGGGCAGAGCGGTGCTGGGAGGGGGAGAAGGGGTCCCCAGGGCTCATG GGCCCTTCGCAGCAGTGGCAGTTGGGGTGGCTGTCTCTAGGGCTGT ACCACGGTGGGTGCCTGGAGAAAGAGGTCCTACCCCTAGTCTTTGCTGCA Contig 45 (300 bp)

TGGGGACCCACTCAGCCCCACTGAGTGACGCCCCCCTGTGGTCCCA CCGCCAACCCTGCCTCACACCAGAGGGGCTGTGGCCACACCTTGTCCACA GCCTGTCCCTGAGACCACGAGCCCCCGGGCTCAGCCCCTCCTCACCCCT GGACCGAGGAGAAGCCCCCACCTGGGCTCAGCTCTTGGAGCTAAACTTCC AGGAAGGTTCTGGTGCCCTCGGGTCTTAGAGCATGGTGGGGAGGGGGATG CTGGTGGGGGCGCAAGCCCTCCCCACATTTCGCACTCGACCCGGTGGGNG Contiq 46 (300 bp)

CCGGCTAGAAGCCACGAGAGCCCCCAGGCCCCGACGTCTCTCCTGC
AGGGATTCGGCAGCCCTGGGGCCACAGGCCTGAGCAGACCTTGGGGTTC
CGGTGTGACTCCAGCCAGGGTCCCTACTGTGTAGGCACCAGGGCAGAGTC
AGCCCTGGGACCATGGCCACAGCTGCTCCCGCCTGAGCCGGCCCCCGC
CCAGGCTGGGCCCCCTCAGTGCACTGTCCCAAGCCAGCTGCTCTCCCCAC
CTCCACCTTCTCCATCCAGGTCCTGCCCCACGGCCTTTGCTCAGGCCCAG
Contig 47 (500 bp)

GGTGGTACATGTGGCCGGAGCCCAGGGCACAGGGTGAGGGAGAAGGGAG CATGCGGGTGCAGACTCGGAGCCCGCGCGTGAGGTGCTGGGTCCTCAGGA CACGCTCTGGGAGTGGAGGACCCCCATCCACGCCCTCACCCAGTGTGTGC CCGCCTGCTCCCCCGGAAACCCTCACAGACACGAGGGCACACCCAGCCCC Contig 54 (1133 bp)

ATGGCGCTCATTAGAATTCGACCTCGGTACCTTGGGATCTTTTGACCCCT ACCTCACGCCATCTACAACATTTACCTCCGAATGAATGAGAGACACCAAA AGCAAATTCATAGAAGAGAAAAAAAGGTAACCTGGACTTTAAAAATGTAA ACTTCTGCTCTTTAAAAGGCAGTGCTAATGAAGTTCAAATACAAACCACA GACCATAAGAAAATACTTGCAAATCTTGTTCTGACAAAGACTAGTGTTCA GAACATACGACGATCAGGGAGAGGAAAACCAGCAATCCTATAAAACTGGA CAAAGAATTGGGGGGAAAAAAAACCCACTTGGCCAAGAAGTTGGTAAATA AGGCCATGAAAACATGCTCAACATCATGAGTCATTAGAAAAATGCAAATT AAAATTATAATGAGATACTACTACACAGCTATTTGAATGGATAAAAAATG TTTTAAAAACTGATTATACCCAGGTTTGGCAAGAACATGAGAAACGAGAT AGAGCTGGGCACTTCCCTCAAAAGTTAAACATACATCCAGGACCTCACAC AGGCTTTCCACCACAGGTGTTTATTCCAGAGACATGAAAGCGCTCATCCA CACAAAGACTCGTAAATGAAGGTTTATAGCACCGTTTGTGGCCCGAACTG AGAAAACCCAAATGACCTTTAACCAGAGAATATCTAAACAAAATATCCAT TCACATTAATCACCCATAAGAAGGAACGGGCTATGGGGACGGGAACCGTA TTGAAGAGGGTCAAAATACATACGCAGCATCAAAGAAGCCTGCCCAAAGG ACACACTGCAGGGTTCCATGGACTGAAACTCGAGAAGGTGAAAACTCG CCAGCAGTGACAGAGCAGGTCCGAGATCAACCTGATGTGGAGGAAAGT GAACCCTCGTGCGTTGTTGGCAGGACTATAAACTGGAGCAGCCCCTACGG ACAACAGTAGCCCGGGCTCCTCTCCTCCATCTCCCTGGGGAGCCTGAGCC TTGAGACGCTGGGGCAAGTGCACGGCATGCTGCCTCACGTGGGGCCCCGG TGAAAACACGTGGCAGCTGGGGAAAGAATCGTA

#### Contig 55 (735 bp)

#### Contig 56 (500 bp)

AGGAAGAACAGGAAACACGGGTTGAGGAGAAAACGGGTGTCTGGCA
GGGGCACGTGCCAACGGTCCACCGGGTGCTGCCGCCTGCGCCC
CAGAGGGGCAGCTCCGCCCCTCGGGCCGCCCCTGCGCTTGTGCTGGC
TCGCGGCTGGGCTCTGCTTGGCTGGTTACAGCTGGGTGCAGCCGCAGGC
TGTGGTGGTGCCCCGGCCCGTCTCCTGCAGCCCCCCACCCGGCCCTCTC
GCCGGCCTGGCCCGGCCACCCTGCAGTCGAGAGAGTCGCCCTGACG
GCTGATTGGTCCACAGCCTCAGATGCAAACCAGCCCCACCTGGAGA
GCCGCCCGGGACACCCTGGTGGAGCAGAACCAGCCCCGGCCCTGGAGA
GCCGCCCGGGACACCCTGGTGGAGCCAGCAGCAGCCCCC
TGGCTCTGGCCGGGGAAACCGGGCTCCCCCCGGGGGCCCC
TGGCTCTGGCCAGCCTTGAATGCTGACGTGAGCGGTGGCCCTATA
Contiq 57 (500 bp)

GACCCCGTGCGTGGGCTCACAGAGTGTGTCCCTCTGTGACCGATCGTC GTGTCCCCGAGGCCCGTTCTGTGGCAGCTGCGTTATGACCGACTACCTTC GAATGCTCAGTGACTGCCGTGCATTGGACACGCAGTCCGCTACCCTTTTC Contig 58 (550 bp)

TGAGGAGCGCAGGCCCAGGCTGAGTGTGCCCAGCTTACACCCCTGGCAG CTTCGTCCCTCCTGGCCCTAACCCCCATCCTACCCCAGCAGCAGGGGCTC CCCCGGTGGGGCCTGGTGAGCGTCTGACTGGGGTTTTGGAGTCAGGTCTGC TCCAGGCTCAGCCCCATCCCCAAGGGTGCCCTGCAGCACTGCTGCCCAC CCCCTAGCGCCCCAGACCTTCGCCCCTCCAGCCTGGATGTACCCACGGA CCCTGAAAAGTGGGGCTGAGCAGGTGCCCTGGCTGGAGTCCCCCTGACTT GGGGCTGGCCAGGCTGCCCTGGAGGGGCTGTGGGGGCACAGCCTGCCCCA TCCTGGCGGGCCACACCCTGCCCTGGGGTTTGGGGCCAAGGCGGGCACG CCCCATGTCAGGCGGGGGGGAACCAGGTAATTACAGCCTGGCAGCCCGCT CCCCAGACCCCCAGCCCGGAGGGCCCCCACCCAGGCTGTGCCACCAAGA CCTGGCATCCAGGGCCCAAAGCAGGTCAAGGGCAGCTGCTACAGATTCTT TTAAGTTGAGACAGAATCGACACATGACAAGTTCCTGGTTTTAGGTACTT CGCTGCCGGGGCCGCCAGTCAGTTTAGTGACCCAGCACACCCCACACAGG TACAATTGCTCTTCTCAAAAGAGGCCCCTGAGAGAGCGCCTGTCTTGGCT CAGGGGTAATGAGCCCAATGGGTATCCATGAGGTTGCGGGTTCCATCCCC GGCCTCGCCGCGTTGGTTA

## Contig 60 (500 bp)

GGCTCAGGAAGCGCAGGGGCAGCGTGTGGGGCGACGGGAACCATGGGGGT CTGTCTTCCCGCCTCTCCTCAAGCCCACCGCCTGCTGCCCACCTCCGAC TCTGCAGCCAGCATGCCGGCTAGAGCCCCTGTGCACCCAGCTGGTGGCCT CTGGCTAAGGGCAGTGCTGGCTGTGGACGCTGTCCCCTCCCCAGCAGCC CAAGGGTCCCATCTGCCAGGCTGGTGGCTGAGGGTCTGCCCTGTGTGGTCC TTGCAAAAACCCCGCCCTCTCCTGCCCCTTGAGGCGTGAGGGAGACGCGG GCTGGGCGGATGCCCTCGGGCACAGCCGCCGCGGTGGCGCCCTGTCGAG GAGGGGGCTCCGACGTGCCCTGACGGCCGCGGGGGAGAGGGTGAG GCCACCTCCTGGCCACGTCCACCCAGCTGCCACGCCGCCTAGCCAGTGGC CCGGGGCCAAGTCAGCAGAGCCAGGCTTCCGACAAGCAGAGGCTGTAGGC Contig 61 (700 bp)

TTTGAAAAATTTTGAGTCAGTGCAGAATTCGCATCTATTCCGCATTCAGG CTCTCCTGTTCTCACCTTGCCTTAGTGCGGATCTTCTATAACCACCACAG TGACGTTTTCAAGGTACTTTATTGAATAATAAGAAAAAAGTGCACACAAT CATGTAGTTAACTTTCTGTGCTCTTTTGCCAGTTTGAAGGGACCCTCTTTT

TGTGCATCCAACCCCAGTGGCCACGGGGGGTGACCCTCGGCCGGTCAGCC
GCCCGCGTCTCCCACGGAACCGGGCCTTGGCCTGAGGCAGAAGGACCCAG
GACTCCATCCCTGCCCCGGACTCTGCCGGAGGGTGCGGTCTGCACAGAGA
CCCTGTGGGGGTGAGGCCGGTCGGGGCTGGGGTTGAGATGGGATGGTCAG
GGCGGCCCCCGCGGGCCTGCAGGAGGCTGGGTGAAGGAGGGGGCCCAGCT
CAGACGCCCCCAAACCTAGCTTGGGAGAGCTGCAGCCCCGCCCCGTCAAT
CGCGACAGCCTGCCCACAGAAGGCATTCAAATGAGAGACAAATATTTGGG
CTTGAAGACTATACCCAGCCACGTCTCTTTGGGAGCCCAAGCTGCTCCCA
GGCCCTCATTTGGGTATTAATTGGTTTTCGTTTAGAGATTTGCATGCTTA
TCAATGGCCACTGGGCGGCTGGGCCTGGATGCGGTCCCAGGCTTTGTATG
Contig 65 (661 bp)

TCCCACGACCTGCCCTCCAGGGCCACATCTGGCGACACCGTCGCAAGAG
TTGGACCGGCCTGGTGTGGCCACAGCCTCAGGCCTTGTCTGGCCGCCCAG
GCCGGCTCCAGGCTCCAAGGAGCTCTGCCTGCCTCCGGAACCCCAGCA
CCCCGGGCCCGCTTCCCCACCAGACCTGTTTTTTCCAGGTCAAGGTCACAG
CTAATTTGGGCTTAAACTGGACAAGGAGGCCTTATCTGGAGCAGGCTGCC
GGCCCTTTGGCCTCTGCCCTGGTGGGGAGGCCTTCCCAGAGGCTGTGT
TGGCGCTGACCGTGCAGCCCTGAGCTTGAACCCGGATAAGGAGGACCCC
ACCTGGGCTGGAGCCAGAGAGCCCTCGTTCCCCAGCTCCGCAGGGTTCTC
ACAGTCCCGCCCTTGCGCTGGGACCTCGCAGCAGTGAAAG
GTCCAGATGCCCTTGAACCCTGGACCTCCGCTGGCTCCCT
ACCCGCACCGAGGACGACACCCTCGCTGCGTGCCGCACAGCAGCTCCCAAGCCCTCGCACGGGTCCCAAGCCCTCGGGGTCCCAAGCCCTCGGGGTCCCAAGCCCTCAAGCCCTCAAGCCCTCAAGCCCTCAAGCCCTCAAGCCCTCAAGCCCCCCAAGATG

#### Contig 66 (500 bp)

TTTGTTGGATGAATGAAATCATGAGAAAGTGATTGGACCGCCCCGTTCGT
CCAGCTGCTTGCCAGCTGCTTTGTAAAGATGACCTCTCACCTTCTCAGAG
GCCTGGCCGGCCCGAGGTGGCAGTCAGCTGAGATGCCATGCTTGTTTTGGC
ACGTGGGAGGCCCCTGTCCACGGCGTGGGTGCCTCTTGTGTCTAATCAGG
GTCAGGGGGAGCACAGGTGCAGGGCACATGTGGGGCCGGGGCCGATGTC
TGGGGAGGCCGGAGGAGCACCTCCCTGGCCAGGCACCAGGACAGGTGATG
GGACAGACCCCAGCGAGACCCTCCCTGGCCAGGCACCAGGACAGGTGATG
GGGGGCCGCCTCCGGGGCGTGTGACAGAAGCCTCTCAGAGGAGGCCCTCC
CACGGTCTCTGGACCATCAAGGGACCGGGGGCGCTGGGCCTGGGGTCAC
ACCCAGCTGGCCGGCCAGCCCGGGTGGGTCGGCCCGGGCAGTTCAC
Contiq 67 (550 bp)

 ${\tt TTTGCATTCAGCTCGTACCCGGGGATCCTTCCCGGGGGCTCTGGGGGTGGG}$ 

ATGTCAGGATAGTAACCTGGGGTGCTGCAGTGACAATGCCAGATCCTTAA CCACTGTGCCACAAGGGAACTCCTTGACCTAGAATCCTATACCCACTGCA AATATATTTCAAAAAAGGTAAAGTCCTGAGCAGAAAAGCAAAAATGGGAT AATTCATTTCTGGAAGACCTTCCTTGTTAAAGGAAGTTTTTTGGACGTGA TGAAGGTAGAAACTCGGAGGCACACAAAGAAGAAGAAGAAGAAGAAGACAC TGGAAACGGAGCAAATAAAGGTAAAAATAAAGTTCATCTCTTTCTCATTT TTTAATTGCTCCAAAAGATAGCTGACCTCTAAAGTAAAAAATAGTGGAAA TGTAGCATATGTCTCTAGCGTAATTTAAAGTATAACTTATAGCAATGATA GCCCAAATAAAGGAGGAATTGAGAATATACAGTTGCTGTTCCCATTGT GGCTCAGCAGTAATGAACCTGGCTAATATCCATGAGGATGCAGGTTCAAT CCCTGGCCTCACTCAGTGGGTTAAAGGATCCAGGGTTGCAGTGAGATGTG ACGTATGTCACAGACGTGGCTCGGATCTGGCATTTCTGTGACTGTGGCTG TGGTGTAGGCCAGCATCTGCACCTCCGATTTGACCCCTAGCCTGGGAACC ACCATATGCTGCTGGTGTGGCCCTAACAGACACAAAATAAAATAAAAATA AAAGAGAGAGAGAATATACCATTGTAAATTTCCTCACATGACACAAAGAG CAATGTGATATTATTTGGTATATGGTGATTGATTCAAGATGTATATCATA ATATTGATTCAAGATGTATATATTCCTTTTCTAAAAAAGAGATTTATACA ATAAGGCAAGAGTGAAAATAAAGTGGAATGCTAAAGAATAGTTAATCCAA AAGAAGGCAGAAAATGGGGAAAAGACATATAACAGATGGAACAAATAAAA AAGAGCTAATGAGATTGTAAAATTTAATCCAAACATACAGATAATCCCAT TAAATTTAAACACTCTCAACACATTGATTAAAAGAAATTGTCAAATTGAA TAAACAAAGCAAGACCCAACTAGATGCAGACTATGAAAAACCCACTTCAT ATAAAGACATGGGTAGGTTTAGAGCAGAATGATGGGGAAACCATGTCACG CAAACATTTGTCAAAATAAAGCTGGTGTGGCTGTATTCATCTCAGACACA GCAGACTTCAGAACAAGAAACACTGCAAAGGATGAAAGAGATACTGCATA ATGATAAAGGGATCAATTTTCCAAGTGCAGGCTCCAAACAACAGAGGTTT Contig 71 (500 bp)

GACGTGCAGTAGCCATGACCTCTACGGCCCCCACTGACCAGCCCGTGTCC TTGTCCCGAGACCGACCCCTAAGCAATAGGATGCAGCAGAAGTGACAGAA CGGCCTCCGCGATGAGGTCGCAGAGGGCTCTGGCTCTGACTCAGGCCCCT CATCCCTCGCTCCTGGAGCAGGGCCAGGTAGGGGCCCCCCAGAGACGC CCTAGAGGAGGTGACGGCCAGCCCGCCCCAGGGAAGGCCTGGGGAC ACCAGGGAACAGAACGGCACAGGCTCCTGGCACAGTCTCCCAGGAGCCCC CTGGTGGCACAGAAATCCTGACCGGCCCAGTGGAGGGGGCTGGGGCGGGG CTCGGGGAGGAGGGCTGGGTGAGGCCGTCTGACTCCTGGCTGAGCGCCG CATACTTGCTGCCTGCCCACGATGCCGGGCCAGGCCTTCCGCACGGACCC AGGCTCACATTCGCCCTACATGCCACTGTGTGGGAGTTTGGGATGGTGTG  ${\tt CCCGCTGGGCCCGGGGGTCAGGGCACGCTTCCCAGAGGAGCGGGTTCCAG}$ AAGGCCCAGGTGGAGAGGCGATAGGAGGGCTCCAGGGCC ACCTGCGAGGACCCTCCTGGGGGGAAGGGAGCGGAGGAGACAGCCGGGT CCCTTAGGCCAAGGCTGAGTTGTGACCGCAGGGAGAGGAGGAGGAGGAGCA CCCACAGCAGGGCAGGGCTGCGGGAGGCTGTGCTGGGTGGCCGGGTGGT GAGGGCCCCTGGACGCCAGAGTCCCTGCTCCAGCTGCCGCCCCGACCCC AGGTCCACCTTCATTTCACAGCCTGGCCCCCGGCCGCTCTGACCGGCCCT GCCCATGCAGGTGTAGCGGGGCAGTGAGGGCCAGGCTCCGAA Contig 74 (450 bp)

GCAGGCCTGGCAGCAGGAAATGATCCAGAAAGTGCCACCTCAGCCCCCA GCCATCTGCCACCCACCTGGAGGCCCTCAGGGGCCCGGGGGGCA GCCGCTATAAAGCCGGCCGGGCCCCCAGCCCTCTGGGACCAG CTGCGTTCCCAGGCCGCCGGCAAGCAGGTCTGTCCCCCTGGGCTCCCGTC AGCTGGGTCTGGGCTGTCCTGCTGGGGCCAGGGCATCTCGGCAGGAGGAC GTGGGCTCCTCTCTCGGAGCCCTTGGGGGGTGAGGCTGCTGCAGGGCTGCA GGTGCCCTTGGGCTGCCTCAACGCCGCCCGGTCCCGCAGGTCCTCACCC CCCGCCATGGGCCCTGTGGACGCCTCCTGCCCCAGGCTGGCCCTTGC TGGCCCCTCTGGAGCACCCCCCCCGGGCCCAAAGCCTTTCATGAACA Contig 75 (1363 bp)

CCTCCAGCTGGGCCGGCAGGGCACCGTGCCCCTCAGGGGACACCACGGG GGGCCACAGTGGCCTCTCCTGCTCCAGGCTCTGCTCCCGCCTGGGGCCCC CTGGGCCGCCCATGGCCAGGGCAAACTCCCAGTGCGGCTGCCCGTC TGGGCAAAGAGGCCGCAGGCCCGCGTGGTCTTAGCAGGCACTGGCGGA TGCCGNTAACTAACCATTTCTTCCGCAGGAGTCCGAATCTGCTCTGACCA CGGGCCCTAAAAATCGCTCCTGGCCCGCAGAGGATCCCCGAACAGCGGGG CTGCCTCCTGCTGCCGGGCCGGCACTCGGCAGGCACGTGCCCTC GTCGTCCCCAGTCTGTCAACCGTCCCGTCGTTACGATCCCCAGAGTCCCA CGCGCGGGCAGCTCTTTCCACACCCCGCACGGCCCCGGAGCTGCCTGGGC ACCCAGATCGCCCTGACGCCTTTGCTCCTAATTCTGCTGAAATACACAT AACGTCTCCTTGAACGTTTGTCCATTTTCACGGGGACAATTCTGTGGCCG TAGGTACACTCCCCTTGGGGCGCAGCCATCGCACCATCCGCTTCCAGGAG GTCCCGTCGTCCCAGATGGACACTGTCCCCACTGATCCCTAATTCCCTGT CCCCCCAGCCCTGCCCTTCCTGTCTCTGTGGCCCTGGCGCCTCCAGGGA GCCCCTGTGCGTGGGATCACAAAACGTGTGTCCCTTTGCGTCCGGTGTGT GTCTCTGAGCATCCGGAGCTTGGGGTGCTTCCACGCTGCGCCTGTGTCAG GACGTCCTTTTGCGGCTGCGCGATGCTCCCCGTGGGGCTGCCCCA CACTGCGCGTGTTCGCTCATCCATCCACTAAGGCTGAGTTACTTTTGGCG GTTGTGAATACTGCTGTGTGAACACGGGCGTGCAAATACCTGCTGGAGGC CATGCTCTTAGGCCTCTCGGGGGGCACACCCAGAGCGGATATGCTCAATA AGGTAATTCTGTGTTTAGCTTTTTGGGGAACCATCAGGCTGGTCTCCAGA GTGACGGAGCATGCGTCGCATTCACAGGAATGGTGCTCGAGGCTTTGAGG TCTCCACCACTCGCTTCCTATTTTCTGTGCGTCACAGCCGTCGGAACGGC TGGGTGGTGCCTCTGTGTGGCTTCAATGTGCTTTTTCTTTTCCTGGCTAT GAGGTTGAGCGTTTTTTATGTACTTGCTGGCCATTCGCAGGGTTTTTGGG GTTTCTTTTTTTTGCCTTTGGGGACGGCGCCCAGAGCGTATAGAAGT

TGACACCTCCAGGCAGGAGGGTGCAGGCTGGGGTCCCAGGTAATGGTGTG
CTGGCCTGTGGGGCGTGGGCTCAGCTCTTAGGATGGTGGGCTGGGCGCCG
ACCCAGCAAGGACAGGGTGATGGCAGGTCGTGGGCTCAGCAAATGAGTGC
CCAGGTTGTGGGGGTGGGCACTTGGGGCTCAGCGAAATGAGTGC
GAGAGGGACGGGGAGGGAGGGGGCCTTGGCCAGCTGGCCCAGATGCCTG
GATGTGAGCACTCACGTGCCCCGGGGTCCACCTCCCCTCCAGTGCCATCT
GGGCAGGAGGCTCCGATGCCTGTCCCTGGGACCCGCTGTCCTGAAATGAG
GTTCACTTGGTGCCTTCCCCAGAGATGCTCGGTCCGGAAGCTGACGAGGC
AGGAGTGCACAAGGGTCTGGGGAAATGGAGCAGAGTGCGGCTGGGGCACA
GAGGCTGCCCCAGCCTGGGAAGATGGGGAGCTTTGCAGGGGTACCCCGC
CAGCTTGTGGGGCCCTGGATACCCAAGGGTGTGAAGAGGCTGAAGAGCGA
Contig 83 (984 bp)

CTGAGCCCAGCTATGTAGATTAGACCCCGGTCCGTCCCAAATTCTTCTCA AAGCTGTCCCGAGATGAGAGATGAGGTTTTCGTGTCCTGTGCTCTCCTCG CTTCCCCTGGGATGTGCCCTAGGGTGGGAGAGGGTGTGTCCCAGGGCTCA GCAGGCGGTCCCATCTTCCCGAGACGGGAGAGATCCCCTCCTTCTCGGCG CCTGTCCCCACGGCCCCCACAGACACCCCCCCCCCGGCATGGCACCCAT GCACCTGCCATCGTGCCCAGTAGGGGATGGGTTTGGCGAGACTGGAGATG GCTGTAGCCAGTGAGACATGCCCTGCCACGTAGCCTGACCCCCTGGGTGT GCTCTGTGAGATCTGGGGACCCCCAGCACCCTAGGGATCATCTTTGCCA GCCTCCTGGGGAGCCTCTCAGAAATGGGGGCCCCCAGAAGGCTGGCAAAG GTGATGGGGAGCGTGGGAAGTCTGGCGGTTGGCGGGGGGGCA GTGCGGGCTGGGGGGGGTGCTCCGGGGTCGGAAGTGGTCCAGCAAGGT TTTGGACACAAAGTCAGGAGGAGGAGGAGGAGGAGACTTGCAGAATTA CAGGTAGAATCAGGAACCCACATCGACGCCAATTGATCTATCCCCCCCTT TTAATCCCTCCTTAGCTTTTTACGCGCTCAACACCAAATTAAACGTACTC CCCACCCACGTAACAGGGGGGGGGGTGACCCGAAGGACGAGGAGCACACG AAGCCACCATCCGTCACCTTGGCGGCACCAGCCGCTGTCCTGCCCTCCGC CCATTTATCGCCCTTGAATTGATTTTTGTTTTGCTCTGTCCCTGTCGCTT CCAAAGATTTCAGGGGAATGAAACGGCTGCCGCC

#### Contig 84 (550 bp)

TGCCCCTGACAACCCTGCCCTGTTAGCCACACTCGCGACTAATAAGGCGA
GAGGTCAGCGGGCAGCCCCACGGGGAGAAAGTGCCTCCGTGCCCCCCACC
CCTGGCTCTGATGGCCCAGCCTGGCACCCCAAGGTGGCCTCGGCCTTCCT
ACCTCCAAGGTCCAGGCGCATGTCCAAGCACCAGCAGAAGCTTCTCCAGG
GTTGGTGCCTCAGGGCAGAAAGCAGGGGTGAGGCTCCCCAAAGGGCC
ACTGGCACCAATGCCCCCAGGCAGCCCCAGCGAAGGGGACAGCCCACCCC
CAGCCCGGGGACGCAGGCCTGAGGGGACATGGGGAACCCAGAGCAGGGCC
AAGGGGAGCAGAGCCCTCCTCCGGGACTTGAAATCTTTCCCGGGGGGCC
CAGGGAGCTGGGGTCTGCAGAGGGCACTTTCAAAATACGGCCCACCCCCA
AATTGCCACGTGGGCCACAGAGCAAGGAGTCGCTGCCAAAGTGGCCTGGC
TTCAGCGCAGGAAGTTCCCCTCCTGGGGCCTCCCCCTATAGGCACAGG
Contig 85 (500 bp)

TGAGCCAGGGCCTGGCCCAGCTAAGCCCCTGGAGCCCTCCCGGCCTGTTT
CCTGCCTCCCATGCTGGCGGAGCTCGGCTTACTGAGCGGGGGCCAGGCCA
GTGTGCGTGTGGAGGTAGATTCCACTCAGCTGGAGGTTGAGGTGGGCAGG
GGGCCGCAGACCCTCAGGCCAGCTCTGGCCGGCCAGGTCCCTGAAGCTCC
CCCGGCTGGCCTCCCCGTCCCTGCCTCTGGCCTTGTCCTGGCCCTTGCCT
GACAAGCTTCTGTGGCTCTGCCTGCAGAGAGACACTGGCTCCCCGCTC
TCGGATGAGGACGGGGCTTTTCTGCACAAGTCCTGCCCCAGAATGTTTGG

GAATCCCGGCATCGAGGCGGGAAGGGGGATGGAGGGATGGGGCCTACCCA CCCCTGCTCCCCACCCAGAATAGCTGGGCGCCCCCATGGGAGGCCGCCC Contig 86 (913 bp)

CTGTTTTCACGTCTTCTGAGGACACCCCAGAAGAGGGGGCTGCAGGCGCC CATGGTGACTCCATGTTCACTGCTGAGGCCTCTGCAGACCGTCTCCCG CAGCAGCCGCACCGTTTCCATGCCACCAACAGCGTGCGAGGCCGCACTG TCCCCACGGCTGTGCAACTGTTTTGAATCTGAGTTATATAAGCAACAGAC GCTCCTTCAAACACACTCACGTGCACACGTGCGCACAGGCGCACAGACAC ACACACGGAGTAATAGGCCTCCCCCCCCCTGAGCCCAGAGGGGGCCT GGGGCCCTGGAGCCTGTGCTTTAGGGCCTTTTAGGAAAGCTGGTGCCTCC CAGAGGGGCCGCCCGAGCGTTGGCTTCCCAAGTCCCCACCAACCCTCGA CAGACTCAAACGTTGGTTTCTTTCGTGCTTTTTGCCCAAGGGATGGGCCCG AGGTGGCCCTGAGGTTTCAGCCCAGCGCCCCAGGCACCCTTTCTCT CCCGGTCCCCGGCCACTTCATGGGACAGCGGGCCTTCCCCCACGTTGTCC CCTGGGTTGTCGTTTTCGTAATGAGACGGAGGCAGGTGCACCTGTCC TGGGGTGAATTCTCTTCTGCAGGAACTCGCTTCCCCGGCGCCTGGTCTGT CTGTTCCTCGGTTGTTGGAACCTCTCGTCACCAGAAAGGGTGGCTCTGAC GTCGCCCTTTCCCTCGTGGCTTTTGCAGTCTGGGTCTTGTCGGGGAACC TGCCCCAAAGAGGGGAGTGACCCCCCACGAGGGAGACGTAGCTCCTGTGG CGACAGCACCGGGGCCCCCAGATTCATGGGGTTCACGCTCACAGTCGCA TGACGCTGCCTTTGGACGAGGGCAGCTCAAGGGAAGCTTGTTTCCTGCCA CGAGCCACAGGCA

#### Contig 87 (650 bp)

GTCTCCGGCAGGGGTGGGCTCTGAACGTCCAGCTCCGCAGACAAATCAGA TTCCCCCGAGCCCTGAGAAAGCCCCCTCCCCAGCCGTCTCCCCACCTG TCGGTGGACAGAGTGACCCCTGCTGACCCCTGCCCGGGCTCCCGCAGGA GGTGCAGGTGTGGGTGTGAGGCTGGCACAGGCTGGCACAGCCTCCCT AGGGATCCACCCTCCAGCCACCTCCTCTCGGGCCAGCCCCACCCCACCC CCGAGCTACAGATGCCTGCGCATTCGCCCCAAGTGTCCTGGACCCTGGAG CCAGGCAGCCCACCCGCTCAGCCTGGCCAGACCCAGCGTTGCCCTTCACG CCCTCCTCCCGCCGGGTCCTCGCGCTCGTCTCCTCAGGTTGGAAGC CCCTTCCCACCTGCCATCTTGCCTGCGCCCAGGATACACGGCTCAACTCA AGGCCTCACTCCCCCCCTCTCCAAGGCTCTGTCCAGGCCCCTCTCTGAC CTGGCACCACCTGCCGCCTCCTGGCAGCCCCAGCAAACCCCCTGCCACAG TCCACGACAGTCCTCTTCTGGCTCTGCCCCCAGGATGCTTCTAGAACTGG GGGGGGGTCCTTCCAGCCCACGCAGCATCCACTGGGCCCTGGGCTCCCT CCCCAGGTGCCCCTCAGAGCTTGCAGCTGGTGCAGACGGCTCTGCTCCGA ACCCATGCTCCCTGCGCCCTTGGACCTGGTGAGATGTTGCAGGTCATTTG GCTGCACCCAAAAGAGTGGCCCCTCAGGGTCCCCCTGCGCCCCTCCATC

#### Contig 90 (350 bp)

GTACTGTAGGGCCTCATTCGAATAGCCTACTAGGTCACAGCTGATCCACA CCTTAGGCCATCACAACTTCCCAGAGGTAGTGCCGCTCCTGTCGTTGAAC AAGACGGTAGTGACTGCTGTGAGAGCTCAGATCTGGTGGGTCACTGACCG AGTGTGGAACCCTGGGGGAAGGCTGTGGGTGTCCCCGGCTGGGTGGCCA TGTCATGTGCCCCTTTCTATCCCTTGGACGAGGCTGGTTCACTCGGCTCT AGAGCCCCAAGCCCCAGCTGCTCTGCCAACCCCCCAAGCCTGAGCCTCAT CAGACCCACCACCCCATCGCCATGGCTACGCAGGACACACCGCTCTCCAC CCCCACCAGCCGCCCCACCTCCCCGAGGTTCCAAAGCTTGA Contig 91 (1464 bp)

TCCAGGACCTGATGCAGCCACGTCGCGAGGCCCCTCCCACGAGGCCC CTTGTTGACCAGCGCTAGGGAAGGGGACCAGGGAGATGCTGAGAACGGGG CCTTCCGAGGGGGCAGGTGGGACTGACTGTGACCCAACACTCCCCACCCC CCTCTCCCGCTCCAGAGGGTGCCAGCCTGGAAGCTGGCAAAGTCCAATCC ACAGGTGGGCTCACGTGGGGGGCCCCCACCTGGTGGGGCCCC AAGCTGCCTCTGGGCGGGGTGGGGGGCTCCCAGCAGGGTCCCATCCAG CTTCTCCCTGGGGAGACTCACAGTTCTGGGAGAAGGGTCCTGACTGCACC GCAGCGCCCGCCCCCCAGACTCACCCAAGTTCTCTCTCTGCATCGG TGACTGGTCTCCGCATTTGCCCAGGCTGGGCATCTGCCCAGAGGATACGT CCAAAGGCAGGGCAAAGCCGGGCCCGTCCCCGGAGCTCCCCACAGGCGC CAGCGGGGTGGAGCGAGGCTGAGCCAAGGTGCACGCGAGGGCCAGAGAAG GCCGAGGCGGGCAGGAGAGAGCGCCAGCCTGGAGGGGGGTGGCTGCC TCCAAGCTGCCCGGGAGGCTGCCTGCCCACCTCCAGGGAGCAAAGCAGGG AGGCTGCAGCTGGCCCGGCCGGCCGCTCTCCAGGACCACGCGTGGCCCAG GCCTCAACGCTCCTCCCACAGCCCAGGAGACCCAGGGCACCGGGTCCATT TACCGCGGGCTCCGGTTTGCCTGCGCCCTGGGATGGACTGTGGGG GCGGGGCGCTGTCTGGGGAGGAGGGGGGTGTCTGAGGCTGGACACCTTGA AGGCAGGTGAGAGTGACAGGTCCGTGCGCAGGAGCCTTCGGCTCTGGATT CTGGCCCTGAGCGAGGGCTGGCTGGCTGGCTGCCGCAGG AGAGTGTGCAGGGAGAGGAGACGGGGTTTGGCCCCGGAGGTGCCGGGGTG GTGCCCTGGAGTGCGGCTGAGCGGGAAGTGGGTGTTGGCGTCTGGAGACG GGGGGTCGTGGGCTTGGGATGGTGACAAGACCCCCCAGGTGGAGGCGGCC GCAGAGGAGGCAGAGAAGCCAGGCCCAACGGCGGGAGGCCTGGG AGTCAGGAGGGACCAGCAGAGCCCTGGGCTCAGTGTCACCGGTCCTGGCA CCTCGCCGACGGATGTCCTGGCCGTGCAGTGGTTGTCCCCTCACCCTGAG CCCTGAGAACCATGCAGGATGCTGGTGTCACAGCAGGAGAGGGCCAGGGC CTGGGGAGGAGTCTTACTGGAAGGCCTTCTCCTTCCGTTTGCAGCAGGCG GGAATGACTGGGGG

Contig 92 (694 bp)

TGGAGCCAGGGCAGGAGCGGTCCCGAGGCCGTGCGTGACCCGGGGGATGGGCGGACCTGGGGGTGGGCTGTGAGCCCAGGCATAGGGACCCCG

CCAGCCCCATCCCCGGCTGGTCCCCCACCACAGAGCCCCCGTTTCCC AGGGGACAGCACAGCCTGCCCCAGGTCTTACATAAAGTCACCTTCTCAG AGCTCCTGTCGCGGCTCAGGGGAATGAATCTGACCAGCATCCATGAGGAC ACAGGTTTGATCCCAGGCCCCGCTCAGCAGGTTAAGGATCTGGCGTTGCC GTGAGCTGTGGTGGAGGTCGCAAGACGTGGCTCAGATCTGGTGTGGCTGT GACTGAGGTGGCGGCCAGCAGCTGCAGCTCTGATTGGACCCCTAGCCTGG TAAATAAAAGAAGTAAACACCCTTCTCTAGCCATAACCACCTGCCTAGG GGCGGAGGCCAGGAAGCGGCACCCCCCGCCCAGGCTGCCCGTGCGCCC CCCACATGGAGGGGCTGGGCTGCGCAGTAACTGCTTTAACTGACGGGAGC TTCGACCAGCAATTCACCAGCGGGCATGCAGCCGGGAAGGGAAGTTATTC GTGTGTAGCTATTAGGCGCCGGAGTGAGGGTGTGCCTCGCCCTGGGCCCA CCCCTGGGGGGGGCATCACAGGGGTTTTGAACACCTGCCCATGAACACG TGTCTCTGAAATCCGGGGAATGCCCACTGCAGGCATGTTCAAAGGGTCAA GACCGGGGCTCTGCCTGAGAAGGACTGGCGAAGGCCAACTACAAAAGCGC ACCCCTCTGTGCAAACCCCCAACCAATGGAACAAAACTCCAGAGGGGCCA Contig 94 (550 bp)

GTTTGCTCTCAGCAGGCCAGGGCCTCCGAGGCCTTAATAGCCCCATAATGA CAGCGCCCGCTCCTGGCATGGGGCCCGCCTGGCATGGGGCAGGGCAGGG CAGAGCAAGCATGCAGCTTCTACCTTCTTCCTGACCTCGTGGCCCCT TCCGAGGCCTCAGGGGGTCCCCCGAGTGGGACCCCAGCCCTGGCTCTCCT CTCCAGAGCCAGGCCCAAGGCTGGGAGTGGCCCAGAGATGAGGGTGCCCG AGCAGGGCACTGCCTTGGCGTCCCCATCCCTGGCGCCTCAGGGCCGTACT GTCCAAAACCAAAAGAAAGCAGTCAGCAAAACTTCTCCCAGCAAGCTGGG GTCAAAGGTCGCTTCCGAGGCGTGATCAGGGTGGCCTTTGCTACTGTCAC CGTGTGCCCTGGGAGAGGCACAGGGACACACACCTCCGAGAACC TGGGGCTTCCAGGGCGTCAGGCTGCCTGGGCCATCCCGGGCCCCTGTGGT CCCAGGATCTGCCGGGACCGTGAGGCCTGCGTCCCACCCTCTGCCTGGGA CAGGCCCCACAGAGCTCACAGCCAGGGGACCGGGGACAGGGCCCCGCCTG GGCCACCTGCCTCCAGCCTCACCCAGCCTGGGCCCCAGGCCTGTGCCTGC GACACCCTGAGTCTCAGGACGGGCGCGGGACAAAGCCGCCCGGCCCCTCC CCCGGCTGGGAGGAGCCCGCGTGGCCCTGACGTGTGGGCCTGTCAGAGC GGAGGCCGGCGGAGGGATCCACGAGCCGAGGCCCGGAGCTGGCCACCC CACCGGTCGATTCCAGGCACTCAGGGATAATTGGGTGTTTAGAAGTCAGG TTAACAGGTGCCCGAACACGCAGGTCTGGGGAGATGCTGAGGTCGCCAAG

GGGGACCAGGCCCAGCCCTCCAGCTCCACGCATACCTGCTAGGAGCTT
GCAACCTGCGAGAGCTTTGTGGACCCCTGCCGGGTGACCCCTGAAGCTG
GCAGCTCTCCTTGGCTCTGCAGCGGCTCTCTACACTACCCCCTCTCCAGC
GGCCTCGGGCCCAGACATCACCCACCCGCAAGGGAAGCAGCATCCA
CCAGCTGGGCCCTTTTCCCCCAGCCTGTGACCGGCCCCGCGCCCCCTCAC
ACCTCTGCGGTCCAAGACCCCTCTCTGGCTGGGCCCTGGTGCTGCCCTTG
CCGTGCACATCTGGGGTCCATACCCCACCAACAGGCCCCACTTTTCTGTC
TCCCAGTGTCCCCTCAGCTGCCCTGATGGGCCCACACCTGCCTCTCTG
CTGCCCCCCTTGACCGCAAAAAGACTGGGGTCCAGGACCCCCTGCCCAT
GACTGCCCTGGAAGACCTCAAGCCTCTCTCTCAATCCTGACCCTTTAAG
GCTCTTGCCACGGAGAAAGCGGCTGGGGTTGGGGGAGGGTGTGGGTCCCA
AAGCAGCTTGCATACTTCTCCTGACTGGGAGCTCATTCCTCCACAGCGTG
Contig 97 (1350 bp)

CCCGCCTTATTTTTAAATTTCCGAAAACAAAAACCACACCTCTCCCGTCC CCGAAATTATTTTGGTATAGTCTTATTCAAAGAAGTCCTGCCACTGAAGC CCACTTGTCCTGTCCCGGGCTGCTTTGGCCAAGGGCCCTGACGGCCCAG GGTGGCTCATTCCCGCATCCCCGCAGAGGCCGCCTTCACATCCCATGCGG GAGCCTGGCTTCCGGCACCCGGCTGTGCCCTCGCTGTGGCCATGGACTGC TTTCGCAGAAGCATAGGGGCCACAACATGGGACAGCCTCGCTCTGCTCGC GCTTTGCTTCAGGCTCTGGTTCCAGGCTGGGCCCTCCTCGGCCCTGCCCG CTGGGTGCCAAGCAGGGCTGGTCCGGCTGTGCCCCCGGGTCTATAGAAGC CTCTGCAGGGCTTCCTACAGCCAGGCTGGGATTCGGCGGCTGCCCGGGAC CCCGCCCCGCTTCTGCTTCAGTGAGGCCCCACCCTGCCTCACTCGCTGA CATTTCCAGAACAGGGGGTTCCAGGAAGCCCTGAGCCTGCAGGGGACTCA GTGACCAGCCGCATCTGAATTTTCCCTCCTTCTGATCTCTGGAGACACGT CTGGCTCAGCCTGGGTGCCCTGAGCTGGGGACCAGGACAGACCTG CAGATGGAGGTCTGAGCCTGGGCAGGGCAGGGCCCAAGGCTCAGGGAGAA ATTGCAGGTGTGAGATCAATGACCGGAGCCTGGATGGGGCCGCCCTGGCC AGGGCAGCTTTCTCCCTGCAGCTCCCTGCCACTGTCCCCCCCAACTCTGG GCTCCTGCTCTGGACCCAGTTGTGTGTTCCCCTCCTCCCAGCCGAGCCAC CCTCCCCATTCTGCCCCCCCAATCCAACACCCTATCGTGGGAACCAGT GGAGCTGAAAGAAGGACCCCCCAAGGGCCCCCCAGCCGCTGTAATCCTTG GGGGCCTCTGCCCAGGTGCCAGGTCTCGGGCAGGAGGGGCCGCGGGCACA GCCGTGGCAGATGCGCCCCCAAGCCTGGGCTCGGAGGAGCCCCGCCCC ACTGACATTTCCAGGCCGCCCGCTGCAGACCCGGCTGGCCGTGATATTTA GACAGGGCTTATTTGCCGTGACTGGTTTTTGATGACTTTGGGGCCCAGGA TGAGCTCAGCCGAGCCCGCGTTGGCCCACCTTGGTCTCAGCTTGGGTTTG ATAATATAACGCGTTCAACTGAACCGCTGACGCCTGCGTGGGCCGAGGCC Contig 98 (1354 bp)

GCTTGCAGTAGTTCATCAGATTGGACGACTCATAAATGTCAAGACATCTA CAAGAAGCTGACCCAAAAACTCACGTGGAAATGCACGTCAACTGGGAGAG TTGAAACAATTTCTAAAAAGAAGAAGGACGTCGTGGGAGGACTCTTCGCG CTCTTTGGTTTCGCTTCACTTTATATTATTAGTTACTGATTTTCCTAAAA GGTTGGGACAGAATAGAAAGCCCAGAAACGGACCCCCGCAAATGTGGTCA ATTGAGTTTGGGCAAGGATGTGAAAGCGGTTCAGTGGAGAAGAGTCTTTT CAAGAATCTCTGGTCCTGGATCCACTGCTCATCCAGGCCCAAGAGTGAA CTTGGCGCACATTTCTCACAGTGTATACAAAAACTGACTCAAAATAATTC ACATACCGTCGTGTAGCGTATGAAGCCATGAAACATCCAGAAGAAAATCT CGGTAACCTCAGGGCATCTGGGGCCTCCACCCTCAGCACCACTGGCCTTG GGGCCAGATACTTACGTGTTCTCCTGTGCACTGTGGGACGTGCAGCCAAA CCCCAACAAGGTGACCATCAGAAATGTCTCCAGACGTCGCCAAATAACTG CCAGAGAGCACAGGAGCCCCTCACTGAGAACCACAGGGTGGGGCAGAGAG ATCTCAGACATGACACGATTAGGGGAAAACAATCTGACACACTGGCTTTG TTAAATTTAAAACTTTTCCCCTGTAAAAGGCAATGGTAAGACATTAAGAG GCGAAGTGGCAGACTGGGAGAAAATATTTGCAAATCATGTATCAGATACG

## Contig 101 (600 bp)

AGTATATCGGGTGAGACTGGGGACCGGTCTGCCGGGAAGCCCCACCATAA AGGCCACGTTGGGCCACAGTCCGGGCCACGTGAGTGTGGGCGGGTCCGCG GGTCTGCTCTTGGAACACCAGGATCTCTAAGAGGTACCAGCCGAGGCCAA GTTCACGTGAGCAAGTGAGCAAATGACTGAATGAGAGCGTGAGCGAATGA GTGAGGGGTGAGTCCGTCCACCACGCAGCCTAGGCTCAGCCAACCGCTGT CCCCGCGTCTCCACTGGTGACCAGAACGGAAAGAGTGGGGAAAGAGTGGT TGTCTCCCACACCCAGTCCCCAACCCCCTGGACGCCCCACCCCTCCAG GGGTGCCGGGCCTGGCCTGTGGGCCCCAGTCTGGAGGCTCTGGCACCTTC CTCATCCGTTCTCCCAGCACCCCAGGTTCGTGCTGAGCCCTCCTGGCCCA CAGGCCTCGGGGACAAAGAGGGCCACCTGGAGGCTCAGGGAGCCTCACCT GCCTCGTGGTCCTGGCGGAGGCGGGTCTGGACATGTGATAGACCGGCCTG GGCTCAGCAGCTCCTGGAAGATGTCAGGGACAGCCTGGGCCACTCTC CCACCAGGAGAACTTATTCCTCGGTGGGGTCCCCCGGGGAAGGGATGGG ATCCCAGCGGGGACCCCAGAGCGTCCAGCACACGGACCTGTCCCTCCAGC CCCTGCCCCACACGGATGCTCACAGCTCAGCCTCGAACACGCACCTGTTG GACTTTGCCTCCTGAGGCTGTCTTCTCAGCCGACGCGGGCCTCCGCTGCA TGGTCTGGAAGCCCAGTGGGACTCGGTGGTGACAGGGAACAGGGGCTCTT GGAGTGGGGTGCCGGGGGAGCCCCGAGGGAGCTGCTTGGGCCTTTGATGG CTGAGTGGGCTGAAGTCAGGCAGGCTCCCCAGGGCTCCCTGACCCCCCC CACCTCAAAAAATCCAGAGCATCCTTTGCTTTGGGTCTGGTGAGGCTCTC TGAGGTCAGACCCTGCGTGGCTGGGCCAGTGGGGCTGGAGCAGGAAGAAA GCAGGACAGCCCCGCCCCTGGCCCAGACTCCCCAAACCCAGCAGGAGAC ACCTGAAACGGGATGGAACCATCCTGAAAAGAGCCACCTCCTCCTTA TGCATCAGCTGCCGGGGTCTGGGGGGCCCGCCCCAGGCCCCAGATGTCCGG GCTGCTCCCGTCTCACATCCAGGGGTTTCTGGGCCCAGGACTCTGTCCCC TGGTGGGGAAGGAAGGGGACAGTCTGGAGACCCCCCGCCCTCCCCATGCG TGGCGCCGGGGACAAAGCCGGCTGGGGTCTCAGGTTTGGGTTCAGAGCA AACGTTGATCTGACCTGGTTCTGAGATGCTCGGCCCGATGCTGCGTTGTC CCGGCCAGCCCACGGAGGGACGCAGGGTGGCTGGCGGGTCTGGGGGGCC CCTGCCGCACCAGAACGTCTGGCTCAGGTTTTTGTCCTCGTGACCCATC ACTAAGGGCCACCCTCTGACCCGGAGCCCTGTCTCCGAGGTGGGAATTGG GGGCTGTCCCTGGCGTCATAGGACCTGGTTGGGGGCATCCAGGGCTGTGT CATGCCCCTCCCCAGAAGACTCTGGGGGGCTGCGGGAGGGTTTCCCCAGCT TCGGGCCAGCCTGGGGAGGCGGAAGGCGCTGGAGGCCTTGCCTGTCCCA GGGAGCATGGCTTCGCTGCAGACTGGGGCCCCGCACACCCAGCCACCACT GGCCGTCTGGAAGCACT

## Contig 103 (650 bp)

ACTGTGCGGCGCCAGCAGGGTCTGGAAGATTTCGCAGCTGGCTCGGGTCA CGATGTGTCCCTGGATGCGCAGATGTGGGTACTTCTTGGACTCCACGGTC Contig 104 (1630 bp)

GGTGTTGTCACTGTGGCTCAGACCCCTGCTGTGGCACAGGGTCCATC CTTAGCCCAGAAACTTGCACATGCCACAGGTGCAGCCAAAAGAAATTCT TACTAATAAGTTGTTCATTTGCCTTTACGTAGAGTGGCATCAAACAGCAA ATTTAAAACACCATCTATCAATACATAGACCGCGGTCAAAGGGAAAGAAC TTTCTATTTCAGCACCTTTAACATGGCTTTGCCCGAATTTGGGACCAGGG TGCTGTGTTTTCATCTCTCCCTGCAGGTGGTCCCCAGATGACCAGGCCGG TCCTGGGCGGAGGAGCCGGACTGTGGATCCAGTTGCTTCCCAAGACAGG CTGACAGGAGAGCAAGGGCCACCCCAACCGAAACCAAAGCCAGAAC GAGCAGAAAGATGCCGTCTTCCAAGTGGGGGCTGGGAGCTTCCTCCCATC CTCCGGAGCCGTGAGGCTGCCCTGGAGCTGGCAGGAGCCACAGAGGACCC GGCTTTGACCGCCCCTCTGGGACCCACAATCAGGACCCTGACTCAGATGC TGAGGGGCCTGGACAACACCCCAGGACCCTGCTGCTTCCCCAGAACCGCT CCGTGGGGCAGGCTTTCCCTTGGGCACCGATGCACCTTGAGGGCAGAGAC GGGGCCCAATAAACGTTTCCAAACCAGTGGGTGAGGGACCCGACCGGCCC GACACGGCAGCCCGGATGCAGGGACTCCGTGCTTGGCCCAGCCTCCCTTG TGGGGACATCCGTTCTCTGATTGGGTGAGTTTCAGCCACAGAGATATTCC CAGGACTACAAAGCTGGGTCCCTTGGGGCACCTGCTGTCACAAAAAGACA AGGCCCTGACCCCAGTAGCCAAGTTCCCCCAGGGGCTCCCCAGGGTCTG GAGTCTCTGTAAACATCCCCCGGCCCCACCCAGCTTTACCCCAAGGCCGA AAGCACCAGCCCCCTGCACCACAGATGAGGCCCCCATGGCTCCCCGACC TAACTTCTGTCTGCAGTTGGCTTTCAGCCTCGGGTGGGGGCAAGGCCTGC ATCTCAGGCTCCCGGGAGAAGTTGCTGCCTCCACAGCAGAGCCAGGGGCC TGCTGACCACCTGGGCCGGGTCGGATCTGGTCTAGAATGCTGCTAAGGTG TTGCCAGGACTCAGGAATGAAGCCATCCCAGGTTTTGAATCCCCGGTCCC ACCACCTTCCACCTCTGACCTCAGGCACCTCGGCTTTCAGAGCTGCCCTT TCTGACTCTGGGACACGGGGCTGTGAGGCGCTCTCGGTGTGTGACAGCTG GCCCTTTCCTCTCAAAAACGCCCGCCCGAGTGACCTCACGGGAGGCAG GGCCAGGAACCCAAACCAGAATCA

#### Contig 105 (1820 bp)

AGTGAGCCCTGCAGGACAGTCTGCTGAGGGGTGTCTGGGCTCCTCAGAGG CTCATGGCCACGGGCACTGGGAGGATAGCAGGTGGACCCCTGCATCCAGG TCCCAGGTCCCAGGCCCCGGACAGGCTTTCTATCTGCAGGAG GGGGGCTCCTGGGGCAGCAGGGATGTGGCTGTGAGGCCTCGTCAGTCTCC CACACACACGCACGCACGCACACACAGAGGCGTGACCAGGGCTGCA GACAGGGCCATGGGAGGACTGCCCGGCAGTGCACCCAGATGGCCACACGG TGGGGCCCTCGTCCCACTTTTGCTGCTGATGCTTCCGCCCAGGCTGCTGG GAGCAAGCACTAGCTTCCCAGGGCTCTGACCAGAGAGGGATGGGAGGGGT CATGGGTCAACAGGCGCCAGGGAATGGGGAATAGGATCTGAGGGGCGGGG GCAAGGGCCCAGGCGAGGCTGCAGTGCCCAGAGCTCCCTGCACCTGCAG GACCAGCCACAGGCCAACAGCTGCAGGCAGAGCAGGGCTGCTCCTGTCCC CAGAAGCTGGCACAGCACATGGGGTCTGACAGCCCCACCCCGGGCCTCCC ACAGAGGGGGGGTCCCCCAAACTCCTCCCCGTCCCACCTCACAGCTCA GCACGCACACATGAATGCACCTGCAAGCACACACTCACACGTAAGCAG CACACACGCACACACTCAAACACGTACATGCAAGCACATGCTGGTCCT TTGTCCCCGTGGAGGGAGGATGGAGGCCCAGCCCGTGGGGAGGGCATGT GGAGTGTTGGGGGGCTGGCTCCAACGCCCTCGCTCAACAGGCACCAACGC TGGACTGAGATAAGCCGGGGCGCTGGCTCCCTTGGGGCCGCTCAGCAGGT TTGACGCCCACACGGTGGCACTGCCTCTTTCAGAAGACGGATGTGGCC ATGCCACCCTCACAGCCTCACCAGTCCCCCCTCAGCTTTAGTGGTGTCCC TGTCACTGTACCCGGGGCCTTCCTTCTTCCAGGGCCAAAAGCGAGTTCAG GGGACAGTGGCGCCCCATAATTACTCACCCAGGGTGCTGTCCTCTGTGG TGGCCTTGAGGCCAAGGTGCTCCCATGGGGGCCCACAGGGCTGGCAGGGT CACTTCCTGAGAGCACCCAGGGCCAGGGCCAGGCCTGGCCGGT

Contig 106 (1500 bp)

TGCCGAATAGAGGTGGAAACCAAGACCCGAAAAAATGTCCACATTTTTCA ATTATTAGAAATTTAGAAAATATTTTACAGGAGTTAAAAGGTATTCCAT TCTGGGGGCGGTGGGCATGCCCACGCATGCAGGCATTCCCCGACCAGC GACTGAACTCGAGCCACGGCAGTCACCATGCTGGATCCTTAACCTGCTGA GCCCCTGGGCAACTCCAGACACTCCATATTCATGTAAACTATTTTTAAC CAAAAAAATGACAAAGCTTTTCAAAACAAAACACATTTCATGGGAAGAGT GGCATTGCTTCACGCCTGGATGGTCGCTGCGGCTTGCGGGACGACGAGGG CCCCGCGGGAGCGCCTCCGCACGGCGCATCAGGACGTGGTGTCCAGGGA AGCGGGGTCACTTCACGGCCTCTCGGGTGCGCGTGGGTTTCCTTTTCGGC ACCACACCCGGACTCAGCACTTGGGGGTTCTTAAACGTGAGAGGCACTGC GGGGCTCGAAGCCACATCACTGACCTCCTCAGACTCTGTTATGTGAAAAC CCATCCGTCCACGAGACCAAAGAGACAGACGAACAAACGCAAGGTGGCGC CTAGGTTGGGCACAGCATGAGGGCAGAGCGGAAACCTTGGCGAAATCCCG GCGAAGCCTGGACGTCGCCAGCTCTTACTTGACGCAAACATAGGGGGATT CAGGAACTCTCTTTACCGCATTTGCAATTAATTTGCTGCAAATCTAAAAT CGTTCCAAGCACAATGCTCACTGCATGGAAAAACCCAGGGGTAGGTCTCG CCCGATCAGGATGTTTTCCCGTGCCCTCTGTGCGGGTGCTGCCCCCTGCG CTGGTCAGTGAGAAGTGTCCCTCCACCGACGACATGAAACTTCCCAGGTC CACGCTCTCTGCTGTCCTGGACGAAAACTCATCTCTGTGAATCTCCCGCC AGCTCCGCGGGAGCCTTCCAGGGCTGGAAGGACGGCCGTCCCGTTCCAGG GGGCAGGTGCACGCTTCCCAAAGCTCCGCGTCCTGCTAGGACGCTCAGAC GGCATCACCCACAAACCCCACGAACTGTTTCCCTCGAGGCGACAGGCTCG CCCTTCTCCGAGAAAGCAGCCCGCACACGTCAGCAAGGGGCCAGCTGCGT TTGTAACTCAAATGGCCACATAGAGTTTGTCCTGGAGGCACGGGGTCTGT CTGGGCCGCACCACTGCACACGCAGAATATGCTGGGACACGCTCCGGGGT CCAGCTTCATGGAATTAATAAAGTTTACTGCTTCACCAAGTACATTCTTA AGTGTAGCTGGCCGCCAGCCTGGGCGTCCGCTCCGAGGCTGCCTCTCTGC CTGGAACCCTTGTGCTGGGGGACCCTCTCTCCAGCCCCACCCCAGCCCCG AGCCCAGGCAACATCCTTCTTGTAAGACACCCGCTACCCTGCCCTCCCGC TTCTCCTTCTGGATCCAATCTCCTCGCTTCTAAGCTCTCTTGAGGCT Contig 107 (550 bp)

TAACCCACTGACCGAGGCCAGGGATCAAACCTGCAACCTCATGCTTCCTA GTCGGTTCGGTAACCACTGCGCCACAACGGGAACTCCTTTGCTTTT TTAGGATTTCACATACACGTGATAACGTGCCGTATTTATCTTTCTCATCT AGTGGCAGGATTTGCTTCTTTTTTTTTTTTTTTTTTTTGTGGCTGAAAATCAG TCCAGGATTATCTTTTTTTTTTTTTTTTTTTTGTTGGAGGACACAGGCTGCGT CCGTGTGACGCTCTGCCGGGGAATACGGGGGCCGATCGCTTTCTGAGCCAG TGTTCTCATTTTCTTGGGAGAAGTACCCGGAGTGGAACGGCTGGGTCGTC CTGCAGTTCTGTGCTGCATTTTTTGAAGACGCTCGGAGCGCTTTCCACAG TGGCTGCACCGACTGACATTCCCACCGAAGTGCACGGATTTCCCCATCCT TTTTCCACGTTTTCCCCGCACTTGCTATTTTTGCCCTGTGGATGTCGGCC TCTCCGTCAGGTGTGAGGGGGGGTCTCCGTGCGGCCCAGGCGAGGAGCGAC CGTGAGCGTCGTTTCACGTTCCTGTTGGGCCACCTGCGTGGCTTCTCCGG AAAAAGGGCTGTTCAGGCTTCTTGCCCATTTCTCAGTCTGATTGTTTGGG GGGTTTGCTGTTGAGTTGTGAGTTCCGCACGTATGGGGGGCATCAACC CTTTATCAGCTATGCGATTGGCAAGTCCGTTCTCCCATGTTCCGCCGGCC GCCTTGGCACGTGTGGGCGGTCTCCTTGGCTCTTCCTTGGTGCAGAAGGC TGTTTTGATGTCAGATGCAAAAATCCATTGCCAGGGTCTGTGCCGAGAAC Contig 110 (306 bp)

#### Contig 111 (800 bp)

GTTTTCCATGATGCACCAGGGGGGGCCGGGACCGCAGCAGGGAAGGCTCCA TCCTGGCTCTGTAAGACCTTGAAAACACCTCATTCCTCTGGTCTTGGCCT GCTCTTCGGTACGCCAAGTTGCTGAGACTGATGTGGGGATCAGTGGGGAG CAGGAATCTTTCTGATTCAGCCGTTTCAAAGTGTCCCAAGCAGAAGCTGT GATGGCAATGCCAAGGCTATCCATGGAGGTGGCTGTGCCAGGGGCCCCAT TTCCTGGGAGCCCATTCCAGGAAAGGAATCTTGTAGCCCCAGGCTCCAGC GAGCTGTGGATGGTAAGCAGGTGGCCCAAGTCCAATTTATGTCTGTGGTC CCAGCAGGGTGCCCAGGAGGCCCCTCGTAACTCTTAAGAATCTTGGTCTG GTCAGCTAAATTGTATGACCATTGTACTGAGCACACATCCCGTTTAAGTA GAATTTTCAAGGATGACTAGGAGTTTGCCACCTGAAGGCAGGAAGGGCAT TCCAGGCAGAGGTACAGAGGTGAGAGGGAGGCTCTGACACTTTGGGCGT GCAGGGGTTTGATGTGACTGCAGCTGGCACACAGTGTATGCCCAGGCCT GGCACGGCTGTGTTGGTGTTTGGAGAGGAAGGGAGAGGTGAGTTGAGCCC AAGGTCTTCCAGGCCAAAAGACTGAAGGTGACCGCGGCTGTCCGGGGCTG GCCCGCAGACCAGGAGGAGCAGGTGGGAGCTGGCTCTTGTTCCGGGGAC Contig 112 (3062 bp)

AGCTTTAGGCTGTTGGTCTAAAGGTCCTGCCTCCTGGAAGAGACACGCCT CTGTCAGCGGACACTGCTAAACCTAAAGGAAGAACTGCCACCTGGTCACG GGACTTCCTAGGCCAACCAACCTACAGGTGACGGCCCGGAGCATCACGAG GAGGTAGGGGACGGGAAGGGATGCATTTGCTGCTCAGCGGATCCACTGGG GCGTTTCTGGAGCCCCACGCCCACACTTTACTGCAAATGCACAAGCCCC AGGCAGCAGGACAAGTCACAGTAGCTCTGGGTTATCCAAGGAGTCAGGGA CCTACCTGGAAGAGTCTAGAACAGGTGACAGAGGAGGAGGAGGATGGTAC CAGCAGTATAGGGAGAATCAGAAATCTGACCCACCCTGGGGGCCTGACTG GGGCTGGGCCACGGGAGTTATGGGCCCCAGGTAGCATCAGAGGCTCCCAG AAGAAGCAGCAGAAGCTCCACCTTAGGTACAGTTCTGGCACCTCCAAGTT GAGAACATGTCCTAGACAGTGCCTGACCCCAACCCAATGGAGTGTCTGGG ACTAGACTAGGCACGCCATTTTGGTCCCAGGTTGCCCCATCTGTACAAAG GGTGTGCGGCCCCAGGGGGACACAATGAGCTCCCATGGGAAGGGTCTTG CGAATCTCCTTAGAAGCAGATGTAAGAGGTGACGTCCAGCTTGTGCCTGG GATGTAGAAGTGGAAAAAGCACCCCTCCCCGACAAGGATGAAAGCAAGA GGCACAAAACAACCTGAAATTCCCAACGCCCCTGGAGATCCTTGGAGAAC TGGGATTCTCCACCTGTAGGGGCACCTGTGAGGAGGCTGTGTGAGCAC CTGCTGACCTGGCACAGAGGATGCCCAATACTAAGAAGCATCAGCTAAAA GTCTCCAGGAATTCCTGGAAGCTGAGGAAGGGCTCAGGAGAGGGTACAGA AGCCCTGGGGCTATAGATATAAGGGACGTGCACACCCACTTGCAGGTCCC CATGGACCCCAGGGACATTCACAGTGATGGGCAAGATTCCCAAAATGCAC CCCTTGTGTGTGGGCCTGGTTCGGTGGGTCAGCAGACACCACACCAAAGG CACAAAGCACACCCTCAGGCTACTCTCCTCCCTCTCCCTTGTGGAACA TGAGCCTTGAGATGCTGGGGCACGTGAAAAACACTGTCACACTTAGGTCC TGGTGAAAACTGACTGCGGCCAGCGGAAAGAATCATAAAGACCCTACACC CACACACAGCCTTAATTACAGCTGTGAGTGGGGCCTGGAGCCCCAAGAATG TCTACACCCATAAGACATAGCGTTAATCAGAAAAACAAGAACAGCCCCAA CCCCACCACCAGGCTGACAACTAACAGGTCATGTTGGAATATCACTGGGA ATGTTCTAGGAGTGTAGAAAGACACCAACTAGGGCATGATGCAAAGAT AATACTTCAGCCTGGGAGTGGATGTGACACAGGGAAAAGCATAAAGTGAT GGCAGAGGACTTTGATGTCAGTGATGGAAGCCACAAAAACTTCTAGCTTA GCTCCATTCCCAACAAGATTGACTGCAAACCCCATGCTAAAACAACAGCA AAAAGAAAGAATCCTCATTTCCAGGCATAAAATTTTTCCCCCAGTCTCTG CTGTCCTCCATAAGATGTCTGATTTCAACAGGAATTACGAGGCTATAAGA AAGGCAAGAAAAACTACACACTGTCAAGAGAAAGCCATCAGAATAACCA GACTCGTAGCACAGACACTGGAATTGTCAGGATATTTTAAATAACCGTGA CAAATACATTAAAGATTCTAATGAGAAGGGGGTAGACATGTAAGATCACA TAGATTTCAGCAAAGAGATGAAACTCGAAGGAAAATTAAATGGGAGCCCT AGAGTGAAAAACACTGTAGCAGAGAAGATGGGTTCATCCGTAAACATGAC ACAGCTTAGGAAAGAATCAGTGAACTTGAAGACAGGGCCACAGAAAATAT ATAAAAGAACAAAGCATCCAAGAGCTGGAGGGTGACACTGAAGAAGAGAG TAATGGATGAGAATTTCACAGAAGCGTTGTCAAGCAACAAACCATACATC CAAGAAGCTCAGAGAACACCAAGCAAGGTAAGTACTGTAAAAAAATAGCC CGAGGTATACCTCATTCAGGCTGCTGAAAATCCATGACAAAAGAAGTCTT GAAAGTAGCCAGAAACAGAAGGCGTGTTCCATTCAGAGGGAAAAGACACC ATTGTTGCCAGAAACCAAATAAACCAGGGCTGAAAGGGTAAAACTTTTTT TTTTTTTTTTTTTTTGGCCATGCCTGTGGCATGTGGAGGTTTCCCGA TCAGGGATCAAC

## Contig 113 (1300 bp)

AAACGGATAAATACAGGTGACCCACAGGCAGAAGCTGAAGTACAAACAGT
TCACAACGGCACCCAAAAAATACCGAAGGCTCAAGGGTAAATCTGACCCC
AGATGAAAGGCCTTCTCACGGAAAATGGCAAAGTGGCGCTGAGAGGCATG
AGAGGTTCGAATAGATGGAGGGCTCCGCCGTTTTCCCGGGTCCGAGGATT
CAGTGACGTCACGACGCCAATTCCTCTGAAACGCCTCTCTAGGTTCAGTG
CAGCCCAGACCCACTGGCAGCCGCCCTCGCTGCAGAGACAGCCCAGCTGG
GTCTTGAGGTTCCTACAGCGAAGCAAAGGGTCTAGAAAAAGCAGACGTCT
CTGGAAAGGGAGAAGCAGCCGATGGATTGGCATACGGCGACAGGAGATTC
CTCGGACAGTGGCACCAGGAGAGGGTGGACAGAGACTGGTGCAACCGAG
CGGGCCCAGGAATAAGTCCACACCCACACGTACCATCTCGTTGTTTATTT
ATTTTTTCCTTTTCAGGGCCACTCCTGGGGCATGGAGGCTCCCCAGCC

AGGAGTCGAATCGGAGCTGCAGCTACAAGCCTACCCCACAGCCACAGCGA CACAGGATCTGAGCCATGTCTGCAGCCTACACCACAGCTCCCGGCAATAT TGGATCCTTAACCCACTGAGCAAGGCCAGGGACTGAACCCACGTGCTCAT GGATACTAGTTGGGTTTGTTACCACTGAGTCACAGTGGGAACTCCTTTAA TTTTAATTTTTGAAGGTTCAGAACTCTTTAATTTTTTAGTGAGGTATAGA TTATATTACGCACCATTTCTTTCTGACTTCGGTGCACGGCTTTTCAACAA ATGGGTGCTGGACCTGCTGGGTGCCTTCTTCAAATGAACCACAAGCCCTC CCTCGCGCCGTATGCAAAATTTAACTCGAGGGGCTCATAGACATAAACGT AAACTCTAAAGCTATAAAATTTCCAGAAGAAAACGTAAGGAAAACCTTTG GGGTCTTGGGCAAAGATTTCTTACCCATGACAGCAAAATTACAATCTACA GAAGAACTGGTGGCCTTTATCGGCATTTAAAACACCTGCCCTTTGAATGA TGCTGTCGCAAAACCGAACATGCAGCAAAACGGATGCAACTAGCAGGTCT CACACTCAGTGACCCACGTCAGAAAGGGAAAGACACGCCACGTGACATCC CTTAGATGCAGAATGTAAAACACGGCCCCCGTGAACCGACCTCAAGAGAG AGACAGACCTACAGACGCAGCAAATTTGGGGTTGCCGAGGGGGATGCCGG Contig 114 (3000 bp)

TGTGAGACCCCTTGGCGGGCCAGGACCCCCCAAGGTGACCGAAGGCCTCA GCGCCCCAGCCGCCCCATCCCCCTCTTTCCCGACACAGGATTTTTTTCC CACCAAGCTCTGTTCCCTTGGTCACGCTCTCACTTGAGCAGCCTCAGGGT CTCCCGGTGCCTGTATCCACGACAGCGTGACCTTCTTGGTGTGTCAACCC AGGACCCCACGCTGGCCAGCCACGCCTTCCCAGAGCACCCCCGCCCATCC TCAGAGTCCAGAAAGGCCCCCATTGACCCCAGAAACCAAAACGCAGA GACTCTGGGACGCCAGCAAGAACGTACACTGACTCCCACCTGCTTCAGGC ACGGAGGCAGGGTTGGTTATGAGCGACCCCGTGGAAGGGCCTTCTTGTC CATCGAGGGCTTCCAGGGGCTCCTAGACGGGGATGAGTGTGGCAACATG TCGCCGCATTACAAAAGACCCTGCAGTGCTGCTGGGATGGGTCCCCCGGC TAGAAAAGCAAAGGATTCCAGCCCAGTCGAGTAGGAGGCGGCCTCGGAGG CTGCAGAGGCGCGGGGGGCGCTGACCACCACTCGGCAAGCCCCGTGTTGG AGGGGACGCCCGGCCCGGCTGCAGCCGGTGCGCCTCCGGATAAGCTCCTA AGAGGCCGCGTGCCCCATGCACGCGCGTGCACACTCGCTGCCCGAGGG TCCTTCAGCACAGACCTTGTGGGGACGGAGGACCTGGCAGGGGTGTGGCT CTGGGGAAGGGGTCTGTCCCAGGAACCCTGTTCTGGATTTGGGGGTGGGC GTGGATATCCCGTCCCAACCTACAGAAGGGAGGGGCTTAAAAAGAGCCCC TTTGGTGTGAGGGGCCAGCAATCCTTTGGCTTTTTCTTGGCCCACTTGGA GCTTGACGTCTGGTCAGTGACTGGGAGCCAGGGGCCAGAGGGGGGGCAGCCG GGCTGAGGCAGGTTCAGGCCAACCATCTCTCGGCCACACTCCCGAGGTCG GGCAGCTACGGGGCCCCCAGAGACACAAGCCCCAGGGGTCCTTCCCCCCC GCCCCCTGCCCCAGATCACCAGGAGACCCAAGCAGCTCTGCCTCCCGTG CCTGAGAAATGCCCCATCTGGGTACCCAAATCACCCTCCCAGAAGGTAGA TCCCAGGGGCGAGGGACTCCGTTTGGGGCACAGACGGAGGCAGAGCGGG CTGATGGATTCTCCCCCGGTTCAGGGATGCTGGCTGCCTGGCCTCCAGGA GCCGGCGGTGCCATCTGATCTGATTAAGGCCTGCAGTCCCAGCTGGGCGG GCACAGCCTGGGGGCTCGGCGGGGGGAAGAAGGCGCTGTCGCCCCAGC CGGTCAGGCTCGCTTTCTCTTCATTTCCTCTCCATTAAAAGTGTCAGAAC CATTTATTGATTTTTTAAATCAGGACGTGCTGTCCGTGACACAGCAAAGT GAACAAAATCAGAGCAAAGAGAGGCCAGGGCTGAAGCCCCAGAGGGCGGC GCCTCCAATCCGGGTTGTGCCCCGGGGCTCCAAGCCCCTTCTTCTTGG GGTCCTGGGCGTAGTGGCCAGGGCAGAATGCACCTGCCGTCATCCTGGGA GGCTTGGCCATCGCTTGTCTCATGACGCACCGTCGTTCCATATC TACGGAAACAGCTTCGCATTAACAGGCAGGGGAGGCGGTTGTTTCTCCTT TATCTGCCCACCATCGGCGCTGGGGCCCACGTGGAGCCCAGCCGGCTGACT TCCCGCTCGCACGCAGGGCACTGATTGCAGGAACGAGGACATCCAGCCCC CGCCTCTCAATGCCCCGGGTGCTGAGAGCATTTCGCCCAAACGGCTTGGG GGCCTGCCCGTGTCTGCCCGTGGCCTCCAGCACCCTCGGCTGCCAGGCTG CTCTGGAGAGGTGCCCGGGGGCCGAGGGCCAGGGGCACCCTGTTCTGCCC CACGTCTCTGTCCTGCTGAAAGTTCCACCAGACGCGTGCTATACCCTG GGAGTCAGGAGGATGGGGGATAGTTGGGGGCTTGACGTCTGTTTCTGAAAA AACACCGTTTTCCCTGAAATATATATGTATTAATTTTTCGTCAAGATAAA ACTGTGTATAGTTTTTCGTGATGAGAAAACGCATCCATCTTCCTTAGAAA GCCTGAAGAGGTACAGGAGCCTATAAAGGACAAGATGACAGATGCCTCTA ACGCACACCAAATGTGCGGTGGCCCCCAGGGGACCGCATAGACGGGGCGG CTCCAGATGGCCACCGTGTGCGAGGGACACGGTTCAGGGTGGCAGAGTAT

TCATGGAAGCCCTTATCACAACCTCGGATCCAAAACCCACTGCGCGAGTC CAGGGATAGAACTCGCATCCCCACAGACCCTATGTTGGGGTCTTAACCAG CTGAGCCACATGGAAACTGGGTAATCTATTTTTAGATGTTCCTAGGGTTT TTGGCCTTGCCTGTACGTGGGGACGCTGCTGGGCCAGGGATCAAACCCGC GCCACAGCTGTGACCCAAGCAGCAGCAGCAGCACCGGATCCTTAAGCA CGAGGCCAGCAGGAGCCCCTGTGTTTAGATTTTGGTGAGGATACTGCGT GGGATTCAGGATATTCACTTTGGGGCTGTTTGGAATTGCCCGTCGCTGTTT AAGCAAAGAAATCCCTTCACTCTGTGTAACTGTGGGGAAATCCTTTAG TCTCTTGAAACCATTGCGTGTGTTTAAGAGTGGTAACTCTGCCACCATAA ATGCCCAGACCAGCGCCTTCCTGAGATCCGCTTTTGTTGCAAATATCTGG TTTGAATGCTTTGATCGCCCGCACCAGACCAGGGTGGGCGGACGCCGCCG GGGACCCGACGTGACCATCGTGCTTCTGTATCCGCCCTTTCTCCGGCACG CGCCCCTGGTTGCCTCTGGCTGCTTTTAGTGGAGGAACTGAAGCCTCGC CACCCAGACCCCGAGACCGCAGGACCCACAATGCTTCAAACACCTGCCCT CTGACTTTTACAGGTCAAGTTCGCCAACGCCGAATTTGCACCGATTGGCT ACAGAGAGCACGGTGGCGCCAAGCCTCCACTTGGAGTTTTATAAGGTCTC CCTCCAGCTCGCAATGAAAATGAGCTGTGATAAGGCAAAGACAAAATTAG TATGAAATCCAGATGCTTCATCTACAATACAATGACCGCGGGATTTGGGT CTGAGCGACTGAAATCAAGGTGGGCTTCCGGAGGGAGGCTGTTAGAGGAA AGGCATTCACGGAGGCTCAGGTCCGAGAGGCTTCCACACCCCTAAGAGGG CTGAGACGCCAAGTAGGGACCAAGCCCCGCAGTCGGGAGAGCTGGGCAGG AAGGAAGTCTGAGGTCACCCCCACCTGGGGAGGAACTGCCTAGAGAAGCG GGGGCGGAAGCAGGGGATGCCCAGTCCCAAGACAGGGACAGGGCGGAAA GGGCTCTCTGCAGGCCCTCAATGCTGCCACAGTGTCCTCGTAAGAGGGAG GCAGAGAGATTGACACCGGGGAGACCACGGGACCACGGAGGTGGAGACC GGGCTGCCCGCGCGTGCCAGTTGCTCCCGAAGCCGGCCCCTCCCCCAGAG CCTTTGGGAAGAGGCGCCAACCTGCAGTTCTGCTACTCGGGGACAGGGAC AGGGACAGCCCCTGGAGCCGCCTCTTAGGGGCAGCATCCCCCAGAACCT TCCTTAACAGACCATCTGGAGAGAGATGGGTCTGGGCTGCAGCTCCTGGA ACTGTTTTGCCCACCCGGCGAGCACCAGTGGGTGCCAGCCTGGGCTGCCC AGCCTCAGGGCCGGGAGGGCTGAGGGCACTGGGGCCCGGCTCTGGGACT CCCCTGCCTGCCCGTGCAGGACAGCCACCTCCCAGCATCTGCTTCCT GCCACCCACATCCCCAGGACCGTCAGCCCAGGCATGCCCCTGGCGTCGGC CACTCACACCACAGGCCAGGAACCCAAGGGGGCAACACAGAAGGGCAGTT GCCATCTGCAGATGGAATGGACAAACTGGGGTCCGTGATGATGGCAGGCT CTGGGCGCCGGGCTGGCAGGGGGCCAGGACTGTGCGGCCATCACAGGA AGGGCATGACGGGGTGAAAGCAAGAGTGGAAACCTCTGCCACCCGCCTGG 

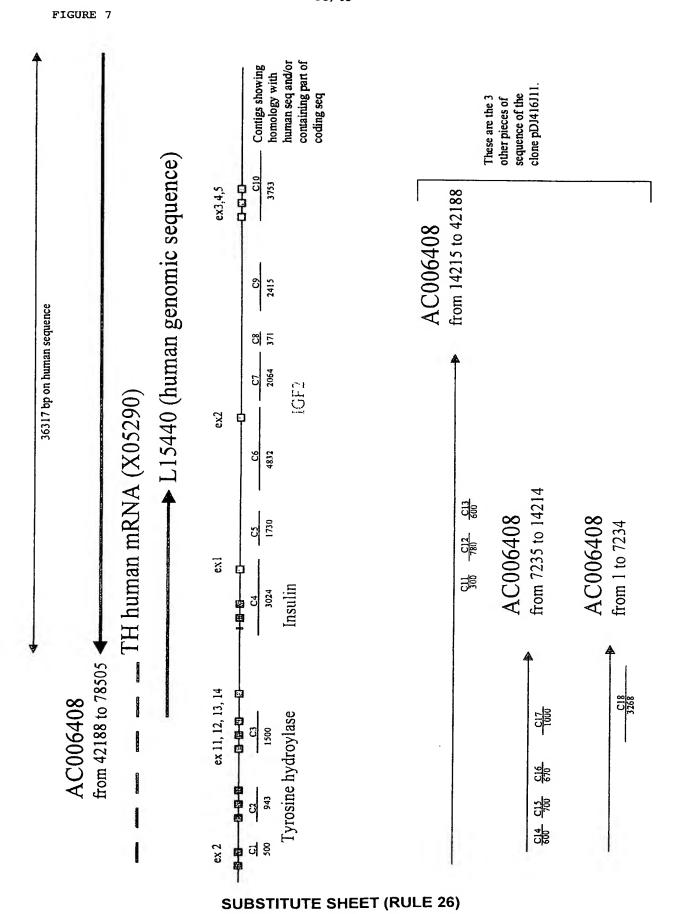


FIGURE 8

## Contig 1 (1040 bp)

GCGCGCCGGATCCTTAATTAAGTCTGAGAGATCTGCGGCCGGGCCAGGGTCTGCTTCTG GCCAAGTGTGGGGCTCTGCTCCATCCTGGCTCGGAGGTCCACCCATGGCAAAGCCTGGGG TCCTCCCACTGAATATTTGGGGGTCCACTCGTGCCAAAGGCTGGGTGTCCAGTGTGCCAA CGGTACATGGAAGCAATGTCTTCCCAAGGACCGTCCAAGGTGTGGTCAGGCCTGGACAGC TGTGAGTCCCTTCGGGACTAGACTTGGTGGCCGAACCCTAGGGACCGTGCCCGAGGGCCC CCACGAGGCCAGGTGTTTGCCCCAGGGACAGAACGGCCAAGGGTGGCCGAGGGTTCTTTT  $\tt CTCTTTGTCCCGATCCTGAGCGGGCAGTGTCCTGGTCGGTGGGGTGCTGGGCAGCCGCAG$ CAGGGCTGAGAGAGCCCGGCTTGTCACTAGGGCGCGCCGGTGAGCCCAGCGGGCATGCCG TGTCCAGACGTTGGATGGGGCAGCGAGGGGACTGGGGTGCCCCAGCCCCCGTGGGAAGCC CGCCCTGTGGAAGCCGCTGTGCTCGCCACAACAAGCACCGTCGACTAGCTGGTGAATCAG CGCCCGTCGCCCGTAATCCCAGGCGCTTTCTGCCCAACCTGAGCCCTGACCCCACACC CCTTGCGACCGCTCCGTGGACCCTGGGCCATCGTG GCACCCAGTGGGGGCTGGGCAGGGAGCCGCCTCCACCTCCGCCCTGAGGGGACGGGACTC GGGGGAGAAGGCCCTCTTTGGAGAATTCCAGGACGGGTGAGGAACGTGTGCTGGACCGGC CGGGTCGGAGGTGGGCCTTG

#### Contig 2 (9234 bp)

GGCAACCAGGGGAAGATGGGGAAGCGGGGTGCAGGGGCGTTTGCGCGGGCCAAGGACCAC CTTGGAAATCTGGAGCCTGGCAGGAGCGGCGCAGGGTTGAGGGGCTTGGCTTGGGCAGGGC TGGCTGGCACCTGGGGGGCTGGCGGGTTGAGGTCCGGGCTCCCAGGTGCCCTATAGGCA GGGCAACATCGGCATGGGGGTGACAGGCCCGAGCTGGGGTGCGGAGGGAAGACGGGGGA GCCAGGCATTCATCCCGGTCAATTTTGGTTTCAGGTCGTGGCGGCTGGTCGGGGGGA GTTGGAGAGAGGTTCGCCCCGGGGCCTGGGGCAGCGGAGGTGTAGCTGGCAGCTGTGGGC TCTGGCCTCCTGCATCCGGAGGTTCTGGGGAGCGAGGGCCGAGGCGAAGCGGCTGAC CAAGCGAGTCTGGTGGGAGACGAGGCAGGGCTGCCAGCAGGGAGACGCAACAGGCGG GGGGCATTCCAGGCCCGGGTCGGACAGGACCCGTCGGGGGTGTCAGGACAGTGGGGTCCC CAGCCGCCACTTCACCCACTGCAATTCATTTAGTAGCAGGTACAGGAGCGGCTCTGGCCG GGCCTCTTGAGGCCTGAGCCTGGAGCCTCGAGGGCCCGGAGAATGGGAAAGAAGGTGCAGTG TGCCAGACAGACGTCACCTGGAGGGAGCACGGCCGTGGGGACGGGCCCCAGAGAGATTTC GGCAGCAGGGAGGCTGCGGGCCCAGCCTGCGGACGTTCCCACGCAGCACTGCGG CCCAGGGCTGCCGCCAGGCCCCCGGTGTCCTTGGTGCACTGTGCGCCCTCGCCGC TCGCCCCTGGGACTGGCACGGCAGACAGGACACCCCAGGGGAGTCAAGGGCACTGACG CTGCGGGTGACAACTGAGCACATATGGGTACCTTTGCGCTCGCACCGGAGACAGGTGAGT GTCTGGCCCCGGCCTGCCGGCCCCGCCACTGCCTCTGCCCTCCGACC AGGGCCCTCTGCTTCCCCACAGCCTCGTCTCCAGTGGGGGTGGACACACTGCCAGCACCA CAGGCCGGACGCCAGGATGTGCTTGGAGGGACATGACACAGTCCGGTGTGACGGAGAGGG ACAGACGTGACGCCGTCCGGCCTTCCTGGTGAGCGCAGGTCCAGGCCTTGGCCCCCAGGC GCCCGGGGGGGGGCTGCCACACCCAGCGTCTGTTCCTTTGCCTTCCTGAAGGAGCACGT GCATGACTGCTCTCTGGACCCCAGAACCCTCAAACGACAAGGTGAGGCAGGTCCCGC CTCGCCCCACACGTGGAAGGGGCGTGGGCGAGAGCCGGGCGCTCACGGTGCCCCCCTCCC CCTGCAGAGATGGTGCTACCCAGCTCATGCCTGGGCCTTGGACCCGGACTTCTTCAAGTC CTCCTAGCTCTGACTCAAGAATATGCTGCATTCTGGAGCCACTACACTACTTGACTCAGG

CTCCCGCTCACGTCCTGTCTCTGTCCTCGCCAGGTTGAGCCAAAGGAACAGACGTC TGGTTTGGAGTCCTGGAACCAGCCCCGGGTCTCGGAGCGGGTGTGTGAGCTGCCGAGTGG CGACCACCGAGCAGACCTCAAACGCTGCACTGAGTGTCCATCTCGTCATGTGCCCCTCCT CGCCAGGGCCACCCCAGAGCCCTGGACTCATCAATAAACTCAGTTACCGGAATCTGTCTC CGGGGGGTAATGAACGTGGGGCTGGGCCCAAAGGGGAGTGGGACGTGGGGATCAGGGCGG GGGGCCTGGAGGATGCAGGGTCCCTGCAGGGAAAGGGGGCCGAGGGCGTGAGGCATGTCC TCAGCCCTGAGAGGCCCTACCCCACAAAGCACAGCCTGCGCGCGACCTCCAGGCCCCCAA CATGCCACCCAGGCTGGCCACACCACTGGGACGCCCATGGGCGGCCACTTTCATCAAGAG CCTGGCAGGCCCTGAGTGCTGGGGCTGGAGGGCACAGAGGGTCCCCCTCCCCTCACGCTTT GCGGTGCTGGGGCACCGCAGGAGTGCCCAACAGGAGACCCCAGGAAGTCTGCTGGGCTGC AGCGAAGGGCAGGTAGGGGGGGGGCCCACAGGGGCCCAGCTCAGTAGGCAGGTGGCAGT GGGAGGCGCAGAAAGTTGGAAAGGGTGGACTGGGCACGTCAGGATCTCGTGGCGGCAGC CCCGGAGCCACGGCCTTGGGTGCACTGCAGCCCCCACGGTTGGTGTCCCGGTCCCAGGCA CCTCCCAGCCCTGGGGCGCCTGGCGTGACGCTGGGAACGCGAGGGAGCAGGCCTCGGAAA CAGGGCTGGGTCCTTGACCCCTTCCTCTGCTCAGGGCAGTCAGGAAATGCCTAGCGGGCC GACTGACCGAGAGAGATAGCGGAGGCCTGGGAGACCCCGCGCTCGTGCCGTTCCCAGCG TGCGTTTGGTGAAGCCCCTGCCCAAAGCACCCTCAGCGTTTCCTCTGCGCGTCCGGCCGC CCCCGGAGGCTTTCCCAAGTCCACGGGCAACTCGCAGGCGAGCCCACTCCACCACA CGCGGGTTTGGCCAGCGCAGAAGCACTCGCCCTTCAGGCGTCAGGAGTTAAGCCCCTCC AAGGCCCGGTGCTAATCAGCTGCCTCTCCTGGAGCTTCGCAAAGCGGGCTCTCAGAGCCC AGCTTCCCGGGGGCTCACCGTGGTGGCATGGGCACCACAGGTGGCCGGAGGGGCACCGAG CACGACGGGGCTGTGGGGGGGGGGGGGGGGGTTGGTGACTCCGAACCTCTACTGAGGC ACACAGAGGACACGGCCGCTTCCAGGGGAGTCAGCCTGCGAAGGGCAGAGGGGCTGTAGC CTCCCGGTCACGCCTCGCCTCGCCCTGGATTCCTCCTGGGGGCCCGCGGCTCGTCGGG CCCTGGGCCCACAGCCCTGTCTTGCCCCACACACAGGGCTGTCTACACTGGGTGCCCACT TGCTCTGCTTCTAGGCTGTTCCCTGGGCAGCTGCCTGGAGGGCCGTGGGCACAGTGCGGG CAGCCAGTGGGGAGGCCGGGGATGGGGCCGGGGATAGGGACCCCTGCCCCTGGGTGAGCC CCACCTGGGCTGGGAAGACAGCAGCGCCCCTTCAGGTCCATGGACCAGGGGACCCAG GGTGGACTGTTTACCTTCAGCCCAGGCCAGTTTCCTGCTTGAGAAAGCCCGGGAGGGG GTGCGGGACAGGCCCGGGCCCCCACGCAAAGGCAGTTTCGCAATGTCCCTGCGCTGACT GGGCACCGGGCTGCTGGGATCTTGGCCCCTGAACCTCCCCGGCCCTGCGGCCAGGGAGG GTTTAGGCTGAGTGACAGCCCACGGAAACCTGGACCCGACATGTCTGTGTGTCCATGTGT GTCTGTGTGTGCGTCCACCTATGCGTCTGCGTGTGTCCATGTGTGTCCACATATCTGT GTCCACGTGTCTGTGTCCACGTGTCTGTGTCCACGTGTGTCCACGTGTGTCCATGTGT CCGTGGACCTGTCTCTTATACACATCTCAACCTG

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FIGURE 8, CONTD.

AAGAGCAACGTCTGAGCTAGCTCCACGCGTGGGTCCATCTCGGCCCAGGTTTAATGAGCC ACTTTCAGGCAGGGATTGCACAGGAGGCAGGGTGGGAAGTGGCTCTGCTCAGACCCCTGA ACAGGGTCTGGAGATTCTCCAAGGGCACAAAAGAACGGACGATGCCCCTGGGGTCAGCGA CAATGCTCCCTGAGAAATCTTGGCACACAGGGCTGGGCCTGCGAGGTGGCCCCTCGCCCC ACCCAGCCTCCTGGAGGACAACCGTCGCCCTGCTCCCAGAGCTGGGGGGGCGCCACACGT GGGGCACAGGAGCATGGGCCCGATTCCAGGCCTGGGCTCCCTCTCGTGTCCAGGATCTC CCCGTGTCTTGTCTCAACAAGCCCCTGACTTGGAGGCCCCAGGGTGACCCCTTAAAGGGG GAACAGAAGGTTCTAGAAGGAGCGTGGCCAGCTTTGGCTTCCCTAGGGCTGTGGTGACCA ACCAGCGGGGCCCCTTCCTGGAAGCCCACCTGCAGGCCGGCTTGCTGGGAAGGGGCCTGC TCCTCGCCGGCCCCACCCGCCCGGGGCCGTTTCCTGGAAGCGGTCACTGGATATTTTGTT CCTTGTCAGCGCCGAGCTTGCATAAAGCAGACACTGAGCTCCTTGTCCTCCGGGAGCACG CGCTCCATCACCGAACACCTGGCCGGACACAGGCGGGCAGCCGGGCCTGGGGGAGCAGCG CGGGCCTGGGGCCGGACCAGAACGATCACGGCGCCGAGCGCAGGGCCCGCGCCCCTTC TGCAGGCCGCCCCACGTGCCCAGGCCCAGCGGTGCCCATCCTGCAGGCTGGGAGGAGGC CTTTGCAGAAACCTTGGCCGGCCTGGATGTCTTGCTGGGAGAGCTGGGGGAGGGGACAGG GCAGGAAGCCGGTCCCCCGAGCGGGGTAGGAAGAGGCCTCGGCCCTGGGAGGAGGAGGA ACAGGACGTGACCTGGGGGCCGGTCCCGGGCCCAGGCGGGCTGGGAGGGCGCCTGGTGG GTCAGCGCCACTCAGAGCCCTGGCAGCAGGGGGGCCTGGGCACGGCTGCAGGACAGAGCTC AGGACACAGATGGGGGCGAGGACTGAGTGGGCCACCACAGATGCTCCCAGGAGGTGGCCA AGGAGTGGCCTTGGGATCCCAGGATGGCCCTGGTCCCAGAAGATGCGGCAGCCCAAGGGA CCAGGCCAGGGCCGCAGGGGCCACAATCTGAGCAGGGCTCAGGCCCAGGGCAGAGGCCC CCTCCCACCCAGCCCTCCCTGGGCCCGCCTCTCC

GTGCAGGCAGTGGGCTCAGATGGGGCAGACATGAGACCAGGTCCAGGGAGAAGCGGGGCC TCAGGAGCACAGACCCCCACCACGGGCTCCCCCAGGTTGGGCGGTGACATCAGCCCTG TGTCAACAGCAGGAGCTGGCAGCTCCCCACCGGGGCTTAGGGAGCGGGGACCCTGAGCCA CCCTGCCACCGCCCCACCGTGGCCCACACGAGGGCCCGCTGCTCTGGGTCTGGGG CCAAGGCCCCCAGGCGCCTGGCACTGTCTCCCCCCTCCCGCTGGCTCTCCAGTG CAAAGCGGACCCCAGGGAGTCCCGCGGAATGTGGGACAGCCCCCCGTAGATCTCGGGGG GGCCAAGCTCTGGTTGACCTCCATCCTGGGGCTGTGGGCCTTTGGTCAGTGGGGAGGGTC ATGACACCCAGCCCAGCTGGTGACAGCCCTGGACGTGCCGGCTCAGGGCTGGCCTGC CCCTGCAGCCTTGAACCCCTGTTCTCTGGGAGTGGGGGCGCAGGGGGGCGCCGGGGCAGGG TGAGAGACGAGAGCCTCTCTTCCCAGAACTTCTGCCTGCGATGAGGACCCAGCAGGGGCC TCTCCTCACCAGAGGGCCTCTGCCGGCTGCAGGGCCCCAGAGAGGCCCAGAGGCTGGAGG CCGGGCCTTGGGAAGAGGCCGGACTTCCAGAAACCAGCTGCCCGCTCCGCAGCACCCAGC GCCCACTTGGGAGGGGGGCGCCCCGTGCCCGGCCCGGGTCCACTGCTGGGGCCGCCA CAATAAAGTTTGTCCCTGCTGGTTACTGTCCGTGTCTGAGAGGTTTCTGGAGCCTGGCCA CAATGGGCGTCAGGATGCGGCTGGGAGGGAGCCTCGCGAGTCAGAGTGTGCTGGTCTCGG GAGGGGTTGGAGAGGGTGGGCGGACGAGGGGCTTCCTGCACTCTGTCCCAGGGAAGCG GGGACCAAGGAGGGGACAGCCCCCGGTCACCAGGAGGGTCCTGTCCCTCTCACCCCCGG GACAGGTGAGCTCCCCGGAGCCGCCCTTCTGGGACAGGACCCCACGGCCAGGCCACGGCC CCCCCACCCGTGGTCCCTCCGTCCCACGGCCGGCCTGGGGGGCCACGGGCCCAGGGCC CCCGCTCCCGTTGGCCCTCCGAGGGTGAACGACCTCGCCTGGGACGTGGGGCAGAGGGC AGGCGCCAAGAGTGACCCCCTGGGACACGTGGCTGTTTGCAGTTCTGGAGGCAGCCGAGA AAGGCCGGGCCGCTCCCCCCCCCCTCTGTGCGGAGGGGGGGCCGGTTGCACAGC AGCCCCTGCCCGCCGCCGCCGCCGCGCGCGCGTGGGACCCGGCCTGGTGCCCCT CCCCGCCCTGCTCAGGGGCCAGCCCTCTCTGGTTCCCAGGACGCCCCGCCCCGCAGG CGGCCAGAGAGTCCCAGAGTGTTAGCCTCCCACGTGTGGGATCCTGTCATATGCGACAGC CTGGGACACTTCAAGGGTTGACATGCTATGCCTGTCACGGATAAATGC

#### Contig 3 (5347 bp)

AGATGTGTATAAGAGACAGGGGCTGGGTGGGAAAGGACAGAGGGTGGGGCCGGAGGAAATG

GGATGCAGAGCCCACCGTGCACGCTCTGCTGGCCTTTGAGCCTCGCTGAGTCGCAAGAAG GGCATGGCTGGAGGGCCCGAGAAGCCACCCAGGCTTCCCGTGCCGAGCTGGGTGCTGGGC CCAGCCGAGCGGGCTTGACGCCACGCTTAGCCCTCCCCAGGGAGCCCAGGGTCGGAAGGA AGAGGCCGGCCGGAGGGCCGTGGCCGCTCAGGCTGGAGGGGGCCCCCGGGTCAGGATGGG CCCCAGACGTCCCCGGCCCATCCGTCACGGAGCTGTCACCCAGGAACGTGCTCC AGACGTGCTTTCCTGCCGCCGAGGCCCCGAGCAGGCTCCAGGCGCCCCCACCCCCGAACG CCCACGCACACCCTCGGTCTGCGAACACCCTGCCGTCATCCGGTGGCCCCGGTTCCCGCC GCCCGCGCCATCCGGGTGCCCCTTCCTCCCTGGGTCGGGGCCATGCCCTCAGCGGGCAC GCAGGCCTGTGCAGGTCTGTTCTGACTCTTCCCCAAAGACGCAGGCCGGCTGCGGGCGCC CCGACCTCGTCTGAGGCCCGTTTGTGCTCACTGGCTGTCTCAGAAAGGGGTGCCCACGGG AAGCGCGTGTTCCTTGGGCCGCAAGGCAAGGGAGCCCACCCCAAGGTGGCTGAGGGCAAA GCCCTGGCTCTCCCCCGGGCAGGTGAGCCCACGGCAGGGGGCTCCCCAGCAGCCTTG GCAGGAAGCAGTGAGGAAGGGTGAGGATGAAGGCAAGGGGGCCTGCGGGGACTTGGGCA AAGCCCCTGAAGAACTGAGTTCCTCGGAAAGGCCGGAGCCCTCAGCCGAGCCTCGGCCTC CGAGCGATGGAGGCGCCCACCTGCGGCCCCAGGGTGCAGCTGTGCATCCGTCCCCCTCG GGCCTCCCCTGCCCCCGGCCACCACACTCTCCCCCTTTTGCCTTTGATCACTTGAGT GCGACAGCTTGTGCGGCCTGAGCCCCAGAGACCGCTGCCCCCTGCCGCCAGCCCACGG GAGCGTCCACCTGGGCCTGGGCACTCATCCCTCCCGGATGAGGCCTTTCTAGCCT CTGGTGGTCTGGGAAGCCCCTGGAACAGGGGGCGCAGGTCCCACACGGGTGCTCTGGCC TCCAGCTGCCAGGGGGGGCCGCGCTCAGGCCAGGGTCCCCTCCACCAGAACCGCCAGGGC CCTGGGGAAAACCTGTCTGTGCTAACAGGGCCGCTCCCCGGGACTCCACGGAGAGGTGCG AGCGGAGGTGCCCGAAGGCGGAAGAGCCCCACCCTCCACTCGGGGACCTATTTCAGCAAGA AGACGGATGGGACTGCCGGGCATGGACAAAGGAACAGGATGAACCTTCTGGAACGCACAA GGCTTCCACGGCTGACCGGTCATAGGAAGGCGCGTCTCTAGGCCAATCCACCGTCCACCG TCCATTCCCCAGCCCTCGAGAGGGGGGCAGGATGGACCGCTGCAGCGTGAGAGAGCTCTGG GGCGCTCCCACAGGGCAAAGTCCCAGGGCACTGACCTCAGAGCCCAACCAGGCCACCGGG GCTGGGCCCACCAGGGAGCCGGGGCCAGGGTCAGGGTCAGGGCCCAGAGTGCGGGAAAGG GTGGCGTGTTGCTTGGGGCGGGGGGGCGCGCAGACGGCCCTCGCACCCCCGACAGCCCT GGAGCTGAGTGAAGCCCGCGGGTCACCTTGGCTGGGGTTGGGGTCTCCTGCGACCGGCAC CCCAGCTCAGGTCATCCTTGCTGTACCGCAGAGGGGCAGGGGTTCTGAGCAGGGACAGGG TGGGCCGCGCAGGAAGCCCCCTTCTCTCTGAGGCTGCCCCGGCCCTGGAGCCTCTCTGGG GCATGCCACCCTCTCACAGACGCCTCCCAGGAGCCCCCACTTTCCTGCTGCGTGAG TTGGAGCAGGTGCAGGGCATCACCACACAGCAGCAGGGCTGTGGGGGCCCCTGAGAGGC GCTCCCAGGTACCCTCCTCAGGGGGGCTGAGCCCGGGGTTGACCCGGGACCTCGCCTGCCC CAAAGCCGGCCCCTCCTCCCGCCCGCCCGACCAGGGCCAGAGAAGCAGGTGTGGGGCGG CACAAACCCAAGTCAGCTTCCAGATCCTGCTGGGGCCGCGTTGAAACTCGAAGCCCCCAG GCTGGGAGGTCTAGACACCCCTGCCCAGACCGACAGCCTGGGCCTGGCTCACAGCTGCCT GGGGGCCCAGGGGTGCACCTGCCCTGTGGGTGGGGGTCAGAGGGCAGGGAACCCTCGGGA AGGTCCCCAGGGTCAAGGTTGGGCCTAAGCTCCGGTGACCTCTGGGAAGTCTGGGGCTG GGTTTTGTTCCCAGAGGAGAGAGGGCCAGTAGCCTCAGAGGGGCTGTGGCACGGTGGGAA GGCCCCAGGTGACCCCAGAGCGTGCGAAGCCAAGCCCCCTTGACTGCAAAGC GCAAAGGGCAGAGGTGGGGTGGGAGCCTCGACCCCCGAGCCCAGGTACACAGGGGGAAG GGCGAGGGATCCGGCAGGGGGCCACACCCCAGGCAGCCCACAAAGCCTTTGGGC CCGGAGCCCCAGATGGGCCCAGCCCAGCTCTGGGAACAGTCTTCCCAGAATTCCCCAGCT GGGATCTCCTAAGTGGCAAGGCCTGTTGGGAGGGGCTGGTGAGAGGCCACTCTGGCGGGA AGACCCCCAGCCACCTGGAGCCCCTAGCCACTGCCTGCGGGCTCCCTAGGGATCCAGG GCCATCAGAGAAGCTCCAGCGACACTGTTTATTTTCAAATGACACTTTTTAAGAAAAACA AGAGCCTGCACGGCCCTCCGGGTGGGGGAGGAGCAGGGGGGCACCCCTGGGACCGGGCCGC AGGCTGTCAGGGCACGGAACGTGTCTCTGGGCCCTGTCCTCAATTCCCGGTGCCCAGTGG CCCCAACTTCCCAGCAGACCCAGCAGGGCCCCAGCTTGTCTTGGCCTGGCCGCTGGTCCT GTCACCCCAGGCCTGGAGTTCTGGAAGATTCTGCTCCTGCTCCCGTGTGCACATACCACT GCAGCCCGCCTGATCTTCCAGGTCCTCCTCCGAGCCCCCGCCTCCAGGAAGCCCTCCAGG AGAGCTCAGGAGGGTCGGCTCCCTGCGCGCAGCTGTCAGACCCCTGGGCCCACCCCGCCG GCTGCTAGGGTCCAGGTTCCCCACAAGCCCTCGGGCAGAGGCTGGGCCGCTGGGTCCCTC GGAGACAACTGGCTCCGAGGCCTTGCCCTAGACGGGTTTCCGGGAGCCCGTCCCCAGCGG

CACCCACTGAGTTTTGAACACTTGGCGCCACCCCCACACCCCAGGCGGTGGCCAGGAGGC CTCCTGGGCAGCAGACAGTCCGTGAGGTGGCCCTGGGGTGGCTCCTGACCTGGGCGCTGG CCCAGCCCTGGGCACAGCTTTCCAGATCTTGCCTGCCGCTTCCTCCAGGCTGCCTCGGCC CCTCCCGCCTGGGGGTGCCCAGCTTTTCCTGGAGGATGCCCACCCTTGCCCATGGTCAGG GAGGGGCTGAGAAACCCCACCTCGTGCCTCTGCCCGGCCTATGCCAGGGGAACCAGGTTC CCTCCCGCAGGAGGGACCGAGTCCCTGACAGCCCACTGCAGAGGGGGAGGAGGTGCCTGG CTCTGCCCCCAGCCCACCAACCCCGTGGCTTCCTGTTTCGCAGCCCACAAAGCACTAAA GGCCGCAGGTCCTGGAACATCAAAGACCCGGGAAGTCCATTGTATTGAATTGAGTGTAAA TGAGCCTGAGGCCTGTGCGTTTCCCACAATTACCGCTGCCCGGGAAGGGCTCCGG AACCGACACAGCCCCCAGGGCCCCTTGCCCATGTGGGGAGCCCAGGCTGGCCTGAAGAAG CCCCATAAGGTGGACCCCACTTTGAGCCCCCACGAGAGTGGGCCAAGGACCAGGTCAGGG GCTGCCCAGGCTCTGGGCCTCCTGCCTGCCAGGTGGGCTCCCTCGGGGCCCAGCCTGG CCTGCAGGACCTTCCCACGCTGAGTTCCCCAGCCTGGTATGAGCGTAGTGGACGGCAGCC ATGCCCAGCACTCAGGGGCCTGAGGGGACAGAGCGGGAACTCCAGCCCCCGGGTCCTCGGC CCCTAGGATCCTTCTAGGTGGGGAAGCCCAAGGGAGCAGAGGGGTGAACGCAGCTGTGTG GGGCCCCAGGCTGCCGAGCAGACCCCTCCTGCTCCACTCCTCGGCCGAGTGGGCGCCGAG ATGCCGGGGCAGTGCCATTTCCCAGGCGCCACCGGAGGCTCCCAGAGGGAGTGAGGCACG GAGGCCCGGAGGGGCCTGGAGTCAATGACCCAGGGATTATCGTGCTGGGTCTTTGCAAA GTTGGCTGAGCAAACGCCGGAGCCAAGGGTCAGGGAGACGGGACTGGCGGGGCCCCGCGG CCCCCTTTCCCCAAAAAAGCCTGTTTCCCAGGTCAAAATCCAGCTCATGATCCG CCCCCTTTGGGACTGATGTTCAGAGGCCCAGTGGTCCCAGCACCTCTGTCCACCGCCCCC CCCACGCTCCCGGGGCCGCCAACCCCTGTGGGCTGCGAGGTGCGGGCACCTCTCCCTTCG AAGCAAAGCCCTGCCTGCGTGGGCAGCGTGATTTCCTGCTTCTCTGGGGCTGCACTTTG ACTGGGGTGGGGGGTGG

## Contig 4 (1592 bp)

AGCCCCTCAGCCCCTCCGAGCAGCTGCTGGGCTCAGCGGGCTCGCCCCCGATGTGCGGC TCCAAGGGCAGGGACAGCTGCTGTTCCGGAGGGTTCCCAGGGGCCCAGCCCACCAGACAG CGGCCTCGGCCCCCTTCCCCGAGGGGCACCCCCACGGAGGGCCCAGACCGGAGGGACTC GGGGCCCAGAGGCCAGGGCAAGAGTGAAGGCAGCGCCGGTGGGAGCGGCGGTCAGCGGGG TCCAGGCTTCAGTTCCCAAGGAGCCCCATGCCCTGAGCCCGCACTGAGCCCTGTGCAGCC TGTGGGTGCCGCCGAGGCCCGCCACCCACCAGCCTGGGGTCGAAGGAGGAG GGGGTGGCCTGACGGATGGTAACAGCTGCTCCCCCCACCTCGCCGGCGTGGACAGGGCTC CCAGCCTCCCTCCTAATCCCCCGCATTTTCCGAATTCTCGGGCCACTGCTTC CTCCTCAAATTCCTGGCCCCCTCGCCCCATCCCCGCCATGGGAAAGGGCCGCGATGCCA GGACACTTGCTCGTCTCGGCCGGGCGGGGGGGGGGGGGCCGGCTGGT GAGGTGCGGGGGTGCCAGGGAGAAGGGCCCAGATTAGGGGGCGTCATGGGAAAGCTGGGA GGGAACGCTACCCAGAGCCCCTCCTGCCGCAGCCTGTGCTGCTCCCTCTCCGCATTTCTG GCCTCTGAGTGCTCCCTGGAGGGAAGGGACCACTGTGTCCTGCCGGCCTCTGGCTCTGCC AGGAATGTCCATCTGTCCGGGCCGGGTTACCTGGCTCAGAGCGTGGGTACCAGCTCATCC AGCCCTGACGCCTGCTCTCGGGAACAGTGGATGGGCCAGGCGCCCCCGTCACACCCCGCA GCTGGGCTCCACAGACGGGCCCGGGATGGCCACGGAGGTGGGGGGGCGCCCCAGGGCGAG CAGGTGCAGCCCGGGGCAGGTGCTGGTGGGGGCTGTGACCCACGTGTGCAGCTCAAGGGT CCAGGAGCCCCAGGGACAGACCTCAGGAGCCACAGCAGGAAGCCTG ACCCCGGGCCTCCTCTCGCACGATTCCCAGGCCAGCCTGGTCTCAGGCAGTCCAAGGTTG  ${\tt CACAATGGTCCCATCGTCCAGAGTTGCAGAGCCAGCACTCTCCCACTGGACGGCGGCCCC}$ TTGACCCTGTCTCTTATACACATCTCAACCCTG

## Contig 5 (831 bp)

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FIGURE 8, CONTD.

ACTCAGCATTTCCCAGGGCACCTGCTGATGGTGCCCAGACCCCGGGGGCCTTCCCGCCGGGCGCGCCCCACGTCGCCCCTCCAGTGGCCACAGCGGGCCTTGGCCAAGGCTGGGAGTTC
TGCACGGGCCTGGGGAGAAGGCGGGGAGAGAGGGGACAGTCTCCTGGCGGGACCAGGG
TGGGGGCAGCAGGTGGGGAGTTCCCACAGCCGGGGCAGCGGGACAGCCGCTTGGCTGCCCT
GGGTCTCAGCCGGGGACAGTGCCCACCAGGAGAGAGAGCGCAGACAGTACAGCCCACCCG
TTTTATATCCTCTCAGGCGGTCTGTGCTTTATTGGGGTAAATATGCAGGACATAGAAACT
CTGCCACTGGACCCCTTGGCCGGGGGACACAGCAGCGGCATTGCATGCTTTCTGGGTGCA
GCGCAGCCAGCACCACCGGCCAGAGCACCCCCATCTTCCCGATCAACCGGAC

#### Contig 6 (4634 bp)

CTCTGGGCTAGCACCGTGGGGGCTTTGCCAGAGTGGAACTGAACTGGGTCCACCCGGAG CCCAGAGGGCGGTGAATGGGAGGCAGAGCCCATCCTGGGAATGGACCAGAAGAAAGGGAG CGGGGGTGGGGAAGGGGCATCAGATCCTGGTCCTTGTCGCCTGCGGTCCCTCTGC CACCACTCCCGAAGCTGATCTGGAGCACACGCGTCGTTAAAGCCGCCATCGAGGCCCCA CTTCTGACAGACGGAAGGGGCAGAGTGCCTTCCTCACCGGCCTCGCCCTGGGAAGGCCC CTCCCTGCAGCCCAGGAAGCCAGCAGCAGGTGACAGAGCCAGGGGCCCCAGGG ACGGGCTCGCGCGCCCGAGCCGGGGGTCCCTTGGCGTCCCCATCCTCTCGTCCTGGAGCC CTCCTGGGTGACCACAGGAATGTGCAAGGCGGCAGCCGGGTGGCGGCCGGGAGGCGGGTG GGAGGCGGGCGGGCCTCTTCACGGGCGGCCTGAGAGATGGGCGCCCGTCCGGCCC TGGCGTCATCGTCTCCGCGTCTCTACCCACTGAGCAAAGACACACGAAATGAAGCTCGAA CGAGCACAGCCAAAGAACGGCCGTTTCTGTCCTTTCTTTAATCCCTTTGGCTTAGGGT GTGACCAATCCCAGGCCACCCAGGCTGTGCCCTGCGTCGTGGGCCATTTCCCAGCCGGCC AGAGATGGAGCAGCCACTGCGGGTCCCCGAGTCTCGGTGAGACAGTCAAGGATGGACCTT GGATGGAGACCGGCGTGCGGCCATGTCCGTGGGTGAAGGAGGCGTGCAGGCCGTGCTGGG GGACATGGTTGCTGTCCCCTCGGCCAAACCATGAAAAGCAGCCCTCTCCCCCAACCCCCA GCACCAACCGGAGACCACCCTCGGCCGGAGCCCAGCACGGCCACCGTCACGTCTCGGTC GTCCAGCTTGGGACAGGTCAGTTCCCAGATGTCCAGGCTGGAGCTGGTCCTTGAAGATCC TAGGGGTCCAGCCCAGCACAGGAGGGCCAGGTGAGAGCCCCCTGTGGTTCTAAGGATGCA  ${\tt ACCAGGGGCCGGGGGTGCCTGCCCTAGAGGGGGTAACTCGGCCCCCTGGGGACCAGTC}$ ACCCCAGGAGGTCCCCAGAGCCCAGCTCGGAGGGCCACAGGTGCCCAGAGTCCCACCTGG GGAAGGCTGCCCCTCCTGCCAGCCCCGAGCCGGGCCCCTGGCGCCCGCGTCCAGCCGCG ACCCCGGGGAGATATTCACCCCCTGCCCCGTGAATCAGGAGGCCCCGAGCCCATGTTTT CAGTCCTTTTCCTCCCATCCCAGCCCCCCAGGAGAAGAGGTGCTGAACTGGGTCCCCTGG AGGCTCCTGAGCCCCAGAACAGTGCCCTCTGAGCAGACGGGCACTCTCAGACCAGCTCAC GCTGGACAAGTCAGCTCCTGCCTGCCGCCTGATGGGCCCTTGGGAGAAGCAGACATGGTG AGGAAAAGGCCCCTGTGCCCTTCACCCTAATTCCCCAGCCCCAAGTCCCACTGGGTTGCC CCTTCCCCGCCCGCCCCCC

ACCCCTGGCCTTGACCTCCAAAAGCACTTGAGGGGGCTTTCTCCAGACACCCTCCAACCC GAGAGGAGTTGGCGTGGACAGCGGGTCAGGCCCCTTTGCCCCGAGGGCAGGGCTGGTG CCACCTGGGTCAGGCGGCAGGCCCTGGAAAAGCACCGGAAATGAGCACACCTGGGTCTCT AGAAGGTTCTTCCAGACCTCTGGGGGCTGAGTCATTTCAACACTCCTGGGCCGGGCAGGG CTTCTTCTTGGCCCCGAGGGACAAGGTCCCCTTCGTCCGGGGGGTACGGCCCCTGGACCC CTGTCCCCGCACCCCACCCTCGCCTGGTGAGGGCCGGCCAGCTCTGGACACAGATC CCTCAGAGCCCCTTCTCCCTCCTCCTCCTCCTCCTCCAAGATGCCCCGGCCTCCAGG TCTGCCCTGACAGCCTCCAAGACGCAGGCACGTCGCTGCGTTCTGCGTCCTCTCA TGGCACAAAACGGTGCCCGCCTAGCTTCCCCCAGAGAAGGGAGATCGTGCTCCCCGGACG GACCCTGCTCTGCCTGCCCCGCCCGGCCTTCAGGGCCTCTCCCCAAGGGTGGCCGCG AGGAGGCCCTCGGCCACGGGGGCTCCATCCTCCCGAGCCCGACAGGCCTCCGCC TGGTGGTCCGACCTCTTCCCCAAGGCCCCGCCCATCCTCCTCGCGCTCCCCCAAACCCTG CCTCTTTCCCCAGCGCCCTTGTCCCCACGGAAGACCCTCCACCCGTGCCATTACACGCTC  $\verb|CCGTCTCCCACGGCAGCAGGGTCAGCAGCTCAGGGGTCCTGGGGCCGTGGAGATGGCC|\\$ TGCCCGGGGGTCTCGCTGACCGCCTCCTACGGAAGCTGTGCCGGGGGGGTGGGGGTGTCTC TGCCCGAACGGCTGGAGGACGAGCCACATCCCAGGGCAGCCGGAACCTGCGTCCT GAGACGGAGAGGCTGGGTGCAGGTGGCTGAGGGGCCTGCACAGCTTGGCCTGGGGTCC  ${\tt CCTAGGTGACACACTGGCTGAACACTCATTGCTGCTCCCCTTCCAGGGTGACCCTGGGG}$ TCCCCGTGTGGCCCTCAGGGCACACGGGGCCCCACCAGGCCTCACAGAACCCCAGTGGG ACTGCACCCAGGGCCCACAGAAGTGCGGGGGCACTGGGGGTCCAGAAACAACCCCACAAC 45/48

FIGURE 8, CONTD.

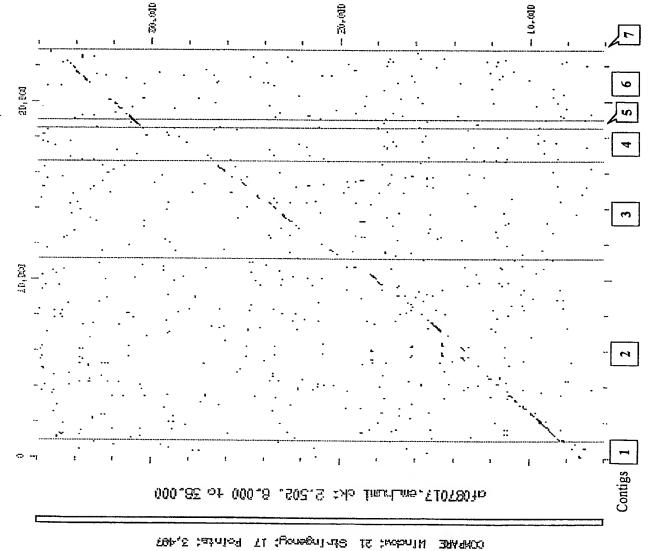
1

CAGGCCAAGGTGGCCAAGGCCTTACTCGAGCGGGGCTGCCCGTCCCAAGAGACTCTGGCC AGTCGTCCGGATCCAGCTTCCCGGGGCCGGGCCGCCGCTGGGCTCCAGGCGGTTCTGGG GGGCCCTCCCCGGGGGTTCGCCCTCCGCTCTCAGCAGCAGGAAGAGGAGCGCGGCCAGC GGATGGGGAGAAGAGGGCCCCTGGCCATCTTGCTCCCCCTGGGACTTGAGGAGGGTCTC GGGCCGGGCAGGCGGGACCCACAGAGACCCTGGAGGAGGCAGCATGGCGGGGAG GTGACCGGGGAAGAGGGCCGTGTCCCAGGCTCACAGCCCGGCCTGGCCGCCCCGGCCCTCG GGAGGCGTGCCGCTGACCGCCTGGCCGGGAGGTTTGCTGCGTGTGGGGTTTGCAGAAAGT CCTCGGGCACTGCTGACCCATCTCCCGTTTCCAGGGCACCAGAGCCACCTAATCTGCCGG CTCTGTGCCCAGGGACAGGCTTGCCTGATCTCTCAAGGCCGGGCGCTCCGCCTTCCCTGG GAGAGGGTTAAACATCCAGCCCAGCCAGCATCTCGGGCAGGTTCCTGGCTCCCCCGCT CGTGCCTCCTCTGAGACCCTGGTCGGCACACCTTTCCCTTGAGAGGAGGAGGAGGAGGAA CGGGCTCCCTGGCTCCGGCTCCGGAAGACAGGGCCGCTCGGCTGGGGCTGCAGGGA GGGGCCCGAGACGCAGGAGAGCCCGGAGGCAAACCCCGCGGGTCTTCCAGAAGGAGG CCTGGCAGGGGGGGGGGGGCCACCACTGCTGTCCCTCTCGTGCCACAGTGGAGGGTGT GGGTGGGCAGTGCCGGGGTGGGAAGTGCAGAAAGACCCTGGACCGTGGGGCTGGGCCGCC CCGGATCACTTCCAGATTTGCTGTGGGACCAAGGGCCGGACCTCGGGGTGACTTCTTTTG TGTGCTGGCCACAGGGGGGCCCCGGCGAGGTCACACGGAAGGGGGCTTCGGACCTGGCCT TCGGGGGACACCGCGGCAGGGCCGGGCAGAGAGGGAGGCAGAGGGAGAGAGGGAGG CAGAGGCCAGAGGGGCCAGAGGGCCACATGCTTGGAGGGCCAGGGAGGAGCGGGA ACGGCGTCCGGCGCCGAATCAGGCCCGTCAGGCGGAGGGTGCGTGGACCTGCC TGGCCTTCACGAGCACAGTCAGCAGGCTGTCTCTTATACACATCTCAACCATCAT

#### Contig 7 (482 bp)

# Human clone at 087017.em\_hum 1: H19 gene + flanking sequences

DOTPLOT of: meq24th.pnt Density; 54094.32 December 6, 1889 12:40



Human clone af087017.em\_huml: H19 gene + flanking sequences

FIGURE 9

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FIGURE 10

**4** ( )

## IDENTIFIED POLYMORPHISMS:

POLYMORPHISMS TYROSINE HYDROXYLASE GENE - CONTIG C3 (figure 6)					
1	GGATCCAGCC (A:T) GCAGCC	1081	bp		
2	ACAACCCCC(-:C)TCCCACAG	1149	bp		
3	TGCGGAGGGG (A:G) GACCTG	1186	bp		
4	AGGT (CAAGGCCAGGT: -) CGAGG	1210	bp		
POLYMORPHISMS INSULIN-IGF2 - CONTIG C4 (figure 6)					
<sub>.</sub> 5	CCC (C:A) CCCC (A:C) CGCCGC	438	bp		
6	CCC (C:A) CCCC (A:C) CGCCGC	443	bp		
7	CGCCGCAGCA (G:A) GCCG	455	bp		
8	GCTTATGG (G:A) GCCGGG	503	bp		
9	CACGGC (T:C) TC (G:A) GAGCA	525	bp		
10	CACGGC (T:C) TC (G:A) GAGCA	528	bp		
11	GTCTGC (A:G) GGCAGGTG	571	bp		
12	CAAGCCCGG (G:T) CGGTT	636	bp		
13	ACCTC (A:G) AGGCCCCCA	710	bp		
14	GC (C:T) GGGCCCAGCCGC	867	рр		
15	ACCAGCTG (C:T) GTTCCC	903	bp		
16	GGC (C:G) CTCTGGGCGCC	1148	bp		
17	GGGGG (C:T) GTCCCGGGA	1305	bp		

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18	GCGGT (C:T) GGGGGAGTT	1320	bp
19	CGCCC(C:T)GGTCCCGCT	1400	qd
20	TCCC (G:A) TCTGCCGGCC	1519	bp
21	GA (T: A) GCCCCATCCCCC	1547	bp
22	GG (C:T) GGCTGCTGCGGC	1607	bp
23	TGGCTGC (G:A) GTCTGGG	2222	bр

## POLYMORPHISMES IN CODING REGION - CONTIG C10 (figure 6)

24	GCGCA (G:T) TGATTGGCA	341	bp
25	CGCCCCCCC(-:C) (G:C)GG	2247	bp
26	CGCCCCCCC(-:C) (G:C)GG	2248	bp
27	GCAGCCGGCTC (C:T) TGG	2257	bp
28	GTTGTTG (C:T) TCTGGGA	2413	bp

## MICROSATELLITES

29	PIGQTL1:	(AT) 11	112	to	133	bp	Contig	57
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30 PIGQTL2: (GT)  $^8$  gcacgcgtgtgcgtgtgtac (GT)  $^{17}$  1074 to 1144 bp Contig

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**31** *PIGQTL3*: (CA) <sup>19</sup> 223 to 260 bp Contig 105